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THE JOURNAL OF HORTICULTURAL SCIENCE

(Formerly The Journal of Pomology and Horticultural Science)

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VOL. XXIV

1948

HEADLEY BROTHERS
109 Kingsway, London, W.C.2
England

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PRINTED AND MADE IN GREAT BRITAIN BY HEADLEY BROTHERS, ASHFORD, KENT

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EXPERIMENTS ON CORRECTING MAGNESIUM DEFICIENCY IN GLASSHOUSE TOMATOES

By **D. J. D. NICHOLAS**
Long Ashton Research Station, Bristol

Magnesium deficiency in tomato has been particularly widespread in recent years in glasshouse holdings in England and reductions in yield have been reported at many centres. In view of this economic loss, experiments were designed to test a number of corrective measures including the use of magnesium sulphate in solution as a foliage spray and either in solid form or in solution as a soil dressing. Progress reports of these experiments have been published for the seasons 1943 to 1946 inclusive (3, 4, 7, 8).

The present paper outlines the experiments and summarizes the main results. The trials have established the superiority of the foliage spray method over soil application of magnesium sulphate both as regards efficiency and economy of material. They have also shown that magnesium sulphate incorporated in a fungicidal spray, i.e. Shirlan A. G., is as effective as when used alone in correcting the deficiency.

EXPERIMENTAL.

General outline of experiments.

Four centres, widely separated and differing in cultural and manurial practices, were chosen for the experiments. Details of location, site, soil characters and degree of Mg deficiency in previous years are given in Table I.

These centres provided good conditions for testing measures for correcting the deficiency in a number of houses. The soil data, given in Table I, show a high available K and P and a relatively low exchangeable Mg content in all soils.

The Mg treatments were usually given in addition to the growers' NPK dressings, but in some of the early experiments K was omitted from the base dressings. Full details of the NPK treatments are given for each centre later.

The magnesium sulphate used was calcined kieserite ($MgSO_4/H_2O$) originally containing 30 per cent. MgO but averaging approximately 20 per cent. MgO during the experiments. The Mg treatments were randomized and were generally applied in duplicate in each experimental house at all centres.

The magnesium sulphate was applied as follows:

In solid form to the soil. Various rates of calcined kieserite were applied to the soil within a week after the grower's NPK base dressing and prior to planting; the rates tested over the experimental period were 5, 6, 10, 15 and 20 cwt. per acre respectively.

In solution to the soil around the roots. A 2 per cent. solution of kieserite was applied to the soil near the roots during the growing season; the dates of application and quantities of salt used were identical with those of comparable foliage spray treatments.

As a foliage spray. Three spray strengths, 1, 2 and 2½ per cent. kieserite were used. A spreader, either sulphonated lorol (1oz. per 10 gall.) or ester salts (1 fluid oz.

per 4 gall.), was included in the spray solution. Sprays were applied at intervals of two to three weeks during the growing season, starting at an early stage of growth. In some experiments, however, comparisons were made between the different effects of sprays applied at early and late stages of growth. The late applications were commenced when symptoms were first seen.

TABLE I.

Data relating to sites and soils at the four experimental centres.

Centre.	Location.	Site.	Soil Characters.	Soil analysis prior to Experiments. Figures represent percentage of the weight of air-dry soil.			Mg deficiency in previous tomato crops.
				Available		Exchange-able	
				K_2O %	P_2O_5 %	MgO %	
A	Lacock, Wilts.	Gentle slope. Southern aspect.	Medium loam, over calcareous gravel.	0·046	0·095	0·009	Serious
B	Langford, Somerset.	Low-lying in wide valley.	Coarse sandy light loam of the Langford series, containing a wash derived from Old Red Sandstone and Carboniferous limestone.	0·035	0·062	0·020	Medium
C	Bath, Somerset.	Low-lying terrace in narrow valley.	Coarse, sandy, medium loam, mainly derived from calcareous alluvial deposits.	0·030	0·071	0·016	Medium
D	Bath, Somerset.	Steep slope. Southern aspect.	Brown, medium to heavy loam derived from oolitic limestone.	0·033	0·068	0·014	Medium

In the latest experiments, the magnesium sulphate was incorporated in the routine fungicidal sprays, i.e. Shirlan, A. G., at the rate of 2 fluid oz. per 4 gallons. These mixed sprays were made up as follows: the magnesium sulphate was dissolved in about 3 gallons of water, then the fungicide, i.e. Shirlan, A.G., suspended in a pint of water, was added and the mixture stirred vigorously, and made up to 4 gallons. Either sulphonated lorol or ester salts, at the rate stated for each, was then added and the mixture well agitated.

The foliage sprays were applied with a stirrup pump, using a three-foot lance fitted with twin, fine spray nozzles.

EXPERIMENTS CARRIED OUT IN 1944.

These were designed to study the effect of soil dressings of magnesium sulphate at the rate of 6 cwt. per acre in the presence and absence of potash in the base dressing.

Centre A. Two houses measuring 39 yards by 7 yards were each divided into four plots, each plot having an area of approximately 68 square yards. The following treatments were given in the form of a base dressing: Plot 1, NP Mg; Plot 2, NP; Plot 3, NPK Mg; Plot 4, NPK. The rates per acre were as follows: N as hoof and horn at 12 cwt. and as nitrate of soda at 4 cwt.; P as superphosphate at 4 cwt.; K as sulphate of potash at 6 cwt.; and Mg as calcined kieserite at 6 cwt. In addition to the above treatment, the two houses received 36 tons per acre of well rotted straw compost. The fertilizer dressings were applied on February 24th, 1944, and the house was planted with the variety E.S.I. on February 27th, 1944.

Centre B. A house measuring 33 yards by 7 yards was divided into four plots each with an area of approximately 58 square yards. The NPK Mg treatments were as for Centre A. but superphosphate was supplied at 2 cwt. per acre. A straw compost at 20 tons per acre was also given. The treatments were applied on February 1st, 1944, and the house was planted with tomatoes (var. Vetomold) on February 4th, 1944.

Centre C. Similar NPK and Mg treatments to those at centres A and B were given in a house measuring 30 yards by 6 yards, so that each of the four plots was 45 square yards. In addition the grower applied a mushroom mould* dressing at the rate of 20 tons per acre. The dressings were applied on February 21st, 1944, and the house planted with the variety Vetomold on February 23rd, 1944.

Centre D. The house was divided into four plots, each measuring 27 yards by 5 yards. Plots 1, 2 and 3 were treated as at centres A, B, C, but Plot 4 was supplied with one foliage spray at the 2½ per cent. rate, in addition to the NPK soil dressing. The base dressings were similar to those of centre A and were applied on February 12th, 1944, and the Mg soil dressings on February 15th, 1944; the house was planted with the variety Potentate on February 27th, 1944. The foliage spray in Plot 4 was applied on April 20th, 1944, at the rate of 150 gallons per acre.

EXPERIMENTS CARRIED OUT IN LATE 1944 (CENTRE D) AND AT THE FOUR CENTRES IN 1945.

The experiments were extended in late 1944 and in 1945 to include a comparison between soil dressings and foliage sprays. Soil dressings included applications of solid kieserite at various rates, viz, 10, 15 and 20 cwt. per acre respectively, and applications of solutions of the material to the soil around the roots. The foliage sprays were usually applied at a 2 per cent. concentration, but at centre D a 1 per cent. spray was also included. The base dressings of NPK and compost material at the four centres were at rates similar to those given in 1944 but K was included in all treatments.

Centre A. A house, similar in dimensions to the one used in 1944, was divided into two blocks A and B and the following amounts of kieserite per acre applied to the soil of the four plots in each block (1) Nil, (2) 10 cwt., (3) 20 cwt., (4) a total of 5 cwt. per acre put on as a 2 per cent. solution on the soil around the roots, twice during the growing season.

* Stable manure compost and soil which had already been used for growing mushrooms.

The Mg was applied to the soil in treatments 2 and 3 on February 17th, 1945, and the house planted with the variety Ailsa Craig on March 21st, 1945. Treatment 4 was applied on March 26th, 1945, and on May 7th, 1945, at the rate of 2½ cwt. per acre on each occasion.

Centre B. A house measuring 33 yards by 7 yards was divided into two blocks, A and B, and the following randomized treatments of calcined kieserite were given to the soil in each block: (1) Nil, (2) 10 cwt. per acre, (3) 16 cwt. per acre. During the previous year Plot 2 had received 10 cwt. per acre of kieserite whereas Plot 3 had had 4 cwt., thus the total for each, for the two years, was 20 cwt. This afforded the opportunity of studying any residual effect due to the previous season's treatment. The Mg was applied on February 15th, 1945, and the house was planted with the variety Vetomold on March 6th, 1945.

Centre C. The same house as was used in 1944 was divided into two blocks, A and B, with four randomized Mg treatments per block: (1) 10 cwt. per acre to the soil, (2) 20 cwt. per acre to the soil, (3) a total of 1 cwt. per acre put on in three sprayings of the foliage with a 1 per cent. solution during the growing season, (4) a total of 2 cwt. per acre in a 2 per cent. solution sprayed on the foliage three times. Treatments 1 and 2 were applied on June 19th, 1945, and the house was planted with the variety Hundredfold on June 22nd, 1945.

Centre D. During the winter of 1944, a large house measuring 147 yards by 7 yards was divided into two blocks, A and B, with seven randomized treatments per block. The Mg was applied to the soil or to the foliage at the following rates per acre: (1) Nil, (2) 5 cwt. to the soil, (3) 10 cwt. to the soil, (4) 15 cwt. to the soil, (5) 20 cwt. to the soil, (6) a total of 5 cwt. per acre to the soil put on as a 2½ per cent. solution around the roots at intervals during the season, (7) a total of 6 cwt. per acre put on in five foliage sprayings with a 2½ per cent. solution applied at fortnightly intervals. The Mg was applied to the soil on September 10th, 1944, and the foliage spray treatment commenced on November 1st, 1944. The house was planted with the variety Potentate on October 12th, 1944.

During 1945 a similar house was divided into two blocks with six treatments in each block. The amounts applied per acre were as follows: (1) Nil, (2) 5 cwt. to the soil, (3) 10 cwt. to the soil, (4) 15 cwt. to the soil, (5) 20 cwt. to the soil, (6) a total of 3¼ cwt. per acre put on in five foliage sprayings with a 2 per cent. solution, at intervals of three weeks during the growing season. The Mg soil treatments were applied on March 13th, 1945, and the foliage spray treatments commenced on May 26th, 1945. The house was planted with the variety Potentate on March 15th, 1945.

EXPERIMENTS CARRIED OUT IN 1946.

These were designed to test the various rates and times of applying foliage sprays. In addition, the experiments included a comparison between the effect of applying magnesium sulphate in the routine fungicide spray, i.e. Shirlan A.G., with that obtained by applying the magnesium salt alone. At all centres duplicate Mg spray treatments were made, but in one block a fungicide, e.g. Shirlan A.G. (2 fluid oz. per 4 gallons) was also included in the sprays. The relative merits of using either sulphonated lorol or ester salts in the spray solutions were also investigated. Soil treatments, usually at the rate of 20 cwt. per acre, were included as a standard to which the results of the various foliage spray treatments could be referred.

At the four centres, houses of similar dimensions to those taken in 1945 were used and the same general scheme of dividing the houses into two blocks A and B with randomized treatments within each block, was adopted.

Centre A. The following Mg treatments were applied in duplicate in the experimental house, (1) Nil, (2) 20 cwt. to the soil, (3) a total of approximately ¾ cwt. per acre put on in three sprayings of the foliage with a 2 per cent. solution during early season, (4) approximately ¾ cwt. per acre put on in one foliage spray, after the appearance of symptoms, (5) a total of approximately 2½ cwt. per acre put on in five sprayings of the foliage with a 2 per cent. solution at intervals of approximately three weeks during the season.

The foliage sprays were applied as follows: Treatment (3), on April 23rd, May 8th, and May 31st, 1946; treatment (4), on June 18th, 1946; treatment (5), on the four dates mentioned previously and, in addition, on June 31st, 1946. The variety grown was Potentate.

Centre B. The following Mg treatments were applied per acre: (1) Nil; (2) 20 cwt. to the soil; (3) a total of approximately 1½ cwt. per acre put on in two sprayings of the foliage with a 2 per cent. solution during the early season and a late one, when symptoms appeared; (4) a total of approximately 3⅛ cwt. per acre put on in five sprayings of the foliage with a 2 per cent. solution at intervals of about three weeks during the season.

Treatment (3) was applied on April 26th, May 17th and June 21st, 1946, and treatment (4) on April 26th, May 17th, June 4th, and July 8th, 1946. The variety Vetomold was planted on March 25th, 1946.

Centre C. The following Mg treatments were given: (1) Nil, (2) 20 cwt. to the soil, (3) a total of 2½ cwt. per acre put on in four sprayings of the foliage with a 2 per cent. solution, at intervals of three weeks, during the season. The variety E.S.1. was planted on April 7th, 1946.

Foliage sprays were applied on April 11th, May 8th, May 23rd and June 18th, 1946.

Centre D. The following Mg treatments were applied per acre: (1) Nil, (2) 10 cwt. to the soil, (3) 20 cwt. to the soil, (4) a total of 1¾ cwt. per acre put on in three sprayings of the foliage with a 2 per cent. solution during the season, (5) a total of 5 cwt. per acre put on in five sprayings of the foliage with a 2 per cent. solution, (6) a total of 1¾ cwt. per acre put on in five sprayings of the foliage with a 1 per cent. solution.

Treatment (4) was applied on June 3rd, June 24th, and July 12th, 1946, and treatments (5) and (6) as for (4) but, in addition, on August 13th and August 25th, 1946.

The variety Ailsa Craig was planted on April 27th, 1946.

METHODS OF ASSESSING RESULTS.

In these experiments it was unfortunately not possible to obtain yield data. At centres A and B, however, the trials were begun because the growers were alarmed at decreases in yields in their houses since the appearance of the trouble. At the former it was estimated that yields had fallen from 60 to 40 tons per acre and at the latter from 50 to 35 tons (decreases of 20 tons and 15 tons respectively per acre). The losses occurred in the later trusses. At centre A it is now known that since the completion of the experiments and as a result of eliminating the chlorosis, the former yield has been restored.

Two complementary methods were used to assess the results of the experiments, namely visual diagnosis (12) and chemical analysis of the leaves. The incidence of the deficiency was recorded by visual means by giving a general severity marking per plot and, in addition, for seasons 1945 and 1946, by determining the percentage of plants affected.

For chemical analysis, random selections of leaves from the mid-stem portions of the plants were made at all sampling times (5, 6). In some instances a tissue test technique was used (5, 6, 9, 11) but, in the main, ash analysis methods were employed for the determination of total amounts of K, Mg, Ca and P. In presenting the data, MgO values as percentage dry matter are given and K_2O values are also recorded where K/Mg interactions are shown.

The apparent Mg content of leaves from spray treatments may be too high in some samples, due to the accumulation of spray residues on leaf surfaces. Leaf samples were taken a fortnight or three weeks after spray treatments, i.e. immediately prior to a further application. A number of mid-stem leaves were periodically tested for spray residues, generally at weekly intervals between spray treatments. These special samples were rinsed for three minutes with an atomised spray of distilled water and the washings collected and tested for nutrients. The results showed that Mg could not usually be detected on the leaf surfaces a fortnight after a spray application. When the growers had carried out top watering treatments, residual spray deposits could not be detected after three to four days.

RESULTS.

SEASON 1944.

In Table II are given the visual and chemical data obtained at the four centres at intervals during the 1944 season.

The Mg and K treatments are clearly reflected in the visual and chemical data and the incidence of Mg deficiency is reduced by the application of Mg, either with or without potash. There is a tendency for the Mg content of the leaves to be greater in the absence of potash but the omission of potash and magnesium simultaneously from the base dressing does not correct the deficiency. The addition of potash in the absence of Mg does not appear to decrease the Mg content of the leaves as compared with those from the plants having no treatment (Nil. Omit Mg and K). The data show that the effect of K in depressing the Mg content of the leaves is not as clear cut as was shown for tomato by Walsh and Clarke (13, 14).

The chemical and visual data also show that the application of 6 cwt. of $MgSO_4$ per acre to the soil is not useful commercially for counteracting the deficiency. In view of this observation the effect of various rates of $MgSO_4$ applied to the soil was tested, late in 1944, and the data are given in Table III. The grower's NPK dressing was applied throughout.

It is clear that all soil treatments reduced the incidence of the deficiency and increased the Mg content of the leaves, as compared with the non-treated plants. A minimum of 10 cwt. per acre is required to bring about a commercial control of the deficiency. The 5 cwt. rate, whether applied in solution to the soil around the roots, or in solid form, is not sufficient to effect a cure.

The outstanding treatment was the 2½ per cent. foliage spray applied five times, at fortnightly intervals, during November and December. This treatment corrected

TABLE II.

Effect of soil dressings on visual symptoms of Mg deficiency and MgO and K_2O content of the leaves at Centres A, B, C and D. Season 1944.

Centre.	Date of Sampling.	Plots and Mg and K Treatments (Mg and K at 6 cwt. per acre respectively).	Visual Symptoms of Mg deficiency.	Chemical data as percentage of the dry matter.	
				MgO	K_2O
A	20/4/44	1. Mg (omit K)	Nil	0·41	4·37
		2. Nil (omit Mg and K)	Nil	0·38	4·55
		3. Mg and K	Nil	0·41	4·74
		4. K (omit Mg)	Nil	0·40	4·77
	19/5/44	1. As above	Slight	0·36	5·74
		2.	Slight—medium	0·31	5·66
		3.	Slight	0·27	5·62
		4.	Slight—medium	0·25	6·60
	26/6/44	1. As above	Slight	0·31	6·24
		2.	Medium	0·23	6·35
		3.	Slight—medium	0·27	7·30
		4.	Medium	0·20	7·01
B	20/4/44	1. As above	Nil	0·59	4·35
		2.	Nil	0·52	4·19
		3.	Nil	0·58	4·49
		4.	Nil	0·54	4·22
	19/5/44	1. As above	Slight	0·45	4·60
		2.	Severe	0·27	4·53
		3.	Very slight	0·37	5·13
		4.	Medium	0·31	5·37
	4/7/44	1. As above	Slight	0·47	5·79
		2.	Severe	0·23	5·64
		3.	Slight	0·47	6·89
		4.	Severe	0·23	6·44
C	19/5/44	1. As above	Nil	0·62	4·55
		2.	Nil	0·49	4·38
		3.	Nil	0·66	5·10
		4.	Nil	0·55	4·79
	14/7/44	1. As above	Trace	0·50	5·91
		2.	Medium	0·31	5·63
		3.	Slight	0·43	6·94
		4.	Medium	0·32	6·53
D	20/4/44	1. Mg	Nil	0·50	3·80
		2. Nil	Nil	0·45	3·94
		3. Mg and K	Nil	0·44	3·81
		4. 2½% Foliage spray of Mg applied once on 20/4/44	Nil	0·39	3·86
	19/5/44	1. As above	Nil	0·48	4·30
		2.	Medium	0·28	4·18
		3.	Slight	0·33	4·50
		4.	Very slight	0·46	4·21
	4/7/44	1. As above	Slight	0·36	4·69
		2.	Medium—severe	0·25	4·43
		3.	Slight	0·30	4·78
		4.	Slight—medium	0·39	4·90

the deficiency completely although the total amount of Mg used was only 6 cwt. per acre.

SEASON 1945.

Further experiments were made to test the effect of various rates of $MgSO_4$ treatments applied to the soil in the presence of the grower's NPK manuring. In addition, foliage spray treatments were included at two of the centres.

TABLE III.

The relation between Mg treatment, visual symptoms and chemical analysis of leaves at intervals during the 1944 season at Centre D.

Date of Sampling.	Plots and Mg (Calcined Kieserite) treatments per acre.	Percentage number of plants showing symptoms and severity marking.		Chemical data as percentage of the dry matter in leaves.			
		BLOCK		BLOCK			
		A	B	A		B	
				MgO	K_2O	MgO	K_2O
14/11/44	1. Nil.	38 medium—severe	49 medium—severe	0·10	4·92	0·15	4·62
	2. 5 cwt.	24 medium	26 medium	0·39	4·67	0·37	4·50
	3. 10 cwt.	7 trace	4 trace	0·51	4·12	0·52	4·22
	4. 15 cwt.	9 trace	1 trace	0·52	4·83	0·60	4·72
	5. 20 cwt.	7 trace	1 trace	0·43	4·68	0·59	4·52
	6. 2½% liquid to roots. (Seasonal total 5 cwt.)	19 slight	25 slight—medium.	0·39	4·95	0·33	4·70
	7. 2½% foliage spray, 5 times at intervals during season. (Seasonal total 6 cwt.)	Nil	Nil	1·75*	3·78	1·63*	3·63
5/12/44	1.	43 severe	59 severe	0·13	5·81	0·13	4·73
	2.	25 medium	29 medium	0·38	5·10	0·34	5·08
	3.	13 slight—medium	9 trace	0·41	5·26	0·47	5·12
	4. As above	16 trace	9 trace	0·62	4·63	0·53	4·91
	5.	15 trace	6 trace	0·42	4·65	0·58	5·1
	6.	25 medium	27 medium	0·26	5·57	0·31	4·81
	7.	Nil	Nil	1·90*	3·76	1·23*	4·01

* Values may be too high due to accumulation of spray residues on leaves.

The data given in Table IV show the relation between treatment, visual symptoms and chemical analysis of leaf samples, taken at intervals during the season.

Visual and chemical data show that soil treatments reduced the incidence of the deficiency at the four centres. The 20 cwt. treatment of the soil is superior to other rates; the leaves usually have a higher Mg content and fewer plants are affected. A minimum of 10 cwt. per acre appears to be necessary to bring about a commercially valuable effect on the deficiency and this confirms the result of a

TABLE IV.

Relation between Mg treatment, visual symptoms and chemical data of leaves at intervals during the 1945 season at Centres A, B, C and D.

Centre.	Date of Sampling.	Plots and Mg (Calcined Kieserite) treatments per acre.	Percentage number of plants showing symptoms and severity marking.		Chemical data as percentage MgO dry matter in leaves.	
			BLOCK A	BLOCK B	BLOCK A	BLOCK B
A	7/5/45	1. Nil.	Nil	Nil	0·61	0·54
		2. 10 cwt.	Nil	Nil	0·87	0·69
		3. 20 cwt.	Nil	Nil	1·08	0·91
		4. Liquid to soil around roots.	Nil	Nil	0·61	0·50
	20/6/45	1.	83 medium—severe	67 medium—severe	0·33	0·40
		2. As above.	37 slight	30 slight	0·50	0·57
		3.	2 trace	2 trace	0·64	0·68
		4.	50 medium	56 medium	0·41	0·42
B	7/5/45	1. Nil	20 medium	30 medium	0·30	0·30
		2. 10 cwt.	6 trace	8 trace	0·46	0·48
		3. 16 cwt.	7 trace	8 trace	0·58	0·54
	19/6/45	1.	95 very severe	100 very severe	0·18	0·21
		2. As above.	64 medium	56 medium	0·30	0·29
		3.	44 medium	47 medium	0·39	0·42
C	16/8/45	1. 10 cwt.	Nil	Nil	0·58	0·53
		2. 20 cwt.	Nil	Nil	0·52	0·59
		3. 1% foliage three times (1 cwt. for season).	Nil	Nil	0·87	0·73
		4. 2% foliage three times 2 cwt. for season).	Nil	Nil	1·23*	1·06*
D	10/5/45	1. Nil	10 trace	9 trace	0·50	0·53
		2. 5 cwt.	1 trace	Nil	0·59	0·69
		3. 10 cwt.	Nil	Nil	0·69	0·66
		4. 15 cwt.	Nil	Nil	0·85	0·62
		5. 20 cwt.	Nil	Nil	0·87	0·85
		6. 2% foliage spray (3½ cwt. per season).	Nil	Nil	1·42*	1·29*
	10/6/45	1.	72 severe	63 medium	0·27	0·36
		2.	35 slight	48 medium	0·30	0·34
		3.	12 very slight	12 very slight	0·45	0·51
		4. As above.	8 very slight	9 very slight	0·48	0·55
		5.	6 very slight	10 slight	0·51	0·56
		6.	Nil	Nil	1·37*	1·27*

* Values may be too high due to accumulation of spray residues on leaves.

winter experiment carried out in 1944 (Table III). The 5 cwt. rate at centre D is insufficient to correct the deficiency and the applications of liquid to the roots at centre A, using a total of 5 cwt., is also inferior to other treatments.

At centre B, although the total application to plots 2 and 3 is 20 cwt. per acre each for the two seasons 1944 and 1945 (see page 4), chemical data show that the 16 cwt. treatment is superior to the 10 cwt. treatment when applied in the same year (1945). There appears to be little residual effect from the previous year's applications.

FOLIAGE SPRAY TREATMENTS.

The superiority of foliage spray treatments both as regards their efficacy and economy of materials is shown in Table III and Table IV at centres C and D. Although the quantities of kieserite in the sprays used have been relatively small, viz. from 1 to 6 cwt. per acre for the season, a spray treatment given at intervals during the season has removed the symptoms completely. The quantities used were

TABLE V.

Relation between Mg treatment, visual symptoms and chemical data of leaves at intervals during the 1946 season at the four Centres.

Centre.	Date of Sampling.	Plots and Mg treatments per acre.	Percentage number of plants showing symptoms and severity marking.		MgO percentage of the dry matter in leaves.	
			BLOCK A	B	BLOCK A	B
A	31/5/46	1. Nil	9 trace	6 trace	0·49	0·45
		2. 20 cwt.	Nil	Nil	0·70	0·66
		3. 2% foliage spray three times early season.	Nil	Nil	0·86	0·92
		4. 2% foliage spray once at onset of symptoms.	Nil	Nil	0·52	0·46
		5. 2% foliage spray five times during season.	Nil	Nil	1·00	1·08
	18/6/46	1. As above	35 slight	37 slight	0·36	0·28
		2.	2 trace	3 trace	0·47	0·42
		3.	2 trace	3 trace	0·52	0·46
		4.	35 trace—slight	26 trace—slight	0·35	0·41
		5.	Nil	Nil	0·96	0·87
	31/6/46	1. As above.	100 slight—medium	100 slight—medium	0·18	0·24
		2.	40 slight	80 slight	0·43	0·38
		3.	20 trace	15 trace	0·53	0·48
		4.	37 trace	23 trace	0·56	0·58
		5.	Nil	Nil	0·95	0·86

Centre.	Date of Sampling.	Plots and Mg treatments per acre.	Percentage number of plants showing symptoms and severity marking.		MgO percentage of the dry matter in leaves.	
			BLOCK		BLOCK	
			A	B	A	B
B	4/6/46	1. Nil	20 slight—medium	23 slight—medium	0·54	0·62
		2. 20 cwt.	5 trace	7 trace	0·78	0·78
		3. 2% early foliage spray twice and a late spray	3 trace	Nil	0·86	0·92
		4. 2% foliage spray five times	Nil	Nil	0·96	1·03
	8/7/46	1.	62 slight—medium	68 medium	0·42	0·36
		2. As above.	30 slight	25 medium	0·44	0·47
		3.	15 very slight	20 very slight	0·56	0·62
		4.	Nil	3 trace	0·87	0·91
C	31/5/46	1. Nil	20 medium	25 medium	0·30	0·36
		2. 20 cwt.	3 trace	7 trace	0·59	0·52
		3. 2% foliage spray five times.	Nil	Nil	1·08	1·12
	18/6/46	1.	46 medium	62 severe	0·33	0·26
		2. As above.	15 slight	18 slight	0·40	0·46
		3.	Nil	Nil	1·06	1·14
D	24/6/46	1. Nil	8 trace	12 trace	0·44	0·46
		2. 10 cwt.	5 trace	1 trace	0·48	0·54
		3. 20 cwt.	Nil	Nil	0·70	0·64
		4. 2% foliage spray three times	Nil	Nil	0·84	0·76
		5. 2% foliage spray five times.	Nil	Nil	0.96	0·95
		6. 1% foliage spray five times.	Nil	Nil	0.95	0·87
	13/8/46	1.	36 slight	42 slight	0·42	0·36
		2.	26 slight (very)	30 slight	0·40	0·48
		3.	10 trace	15 trace	0·50	0·48
		4. As above.	5 trace	10 trace	0·66	0·60
		5.	Nil	Nil	0·91	0·93
		6.	Nil	Nil	0·63	0·73
	25/8/47	1.	52 medium	63 medium	0·36	0·32
		2.	30 slight	36 medium	0·42	0·44
		3.	20 slight	25 slight	0·45	0·49
		4. As above.	10 very slight	7 very slight	0·53	0·58
		5.	Nil	Nil	0·84	0·88
		6.	2 trace	3 trace	0·58	0·58

dependent on the amount of the deficiency. Chemical data corroborate this result but the Mg values may be too high due to the presence of spray residues on the leaf surfaces. That these high levels exist within the sprayed plants is shown by (1) the absence of Mg in leaf washing experiments mentioned previously, (2) the absence of symptoms in sprayed plants and reappearance of symptoms when spraying was discontinued, accompanied by values below 0·5 per cent. MgO, (3) a depression usually recorded in the K content of leaves sprayed with Mg as compared with those taken from Mg soil treatments, as is shown in Table III.

TABLE VI.

Statistical analysis of chemical data given in Table V.

Centre.	Plots and Treatment.	Treatment means.	Diff. required for significance between treatments.		Diff. required for significance between different sampling dates.	
			5% level.	1% level.	5% level.	1% level.
A	1. Nil.	0·34	0·04	0·05	0·11	0·15
	2. 20 cwt.	0·51				
	3. 2% foliage spray three times early season.	0·64				
	4. 2% foliage spray once at onset of symptoms.	0·49				
	5. 2% foliage spray five times during season.	0·94				
B	1. Nil	0·48	0·13	0·19	Not significant.	
	2. 20 cwt.	0·58				
	3. 2% early foliage spray twice and a late spray.	0·74				
	4. 2% foliage spray five times.	0·94				
C	1. Nil.	0·31	0·27	Not significant.	Not significant.	
	2. 20 cwt.	0·49				
	3. 2% foliage spray five times.	1·10				
D	1. Nil.	0·39	0·08	0·10	0·10	0·14
	2. 10 cwt.	0·46				
	3. 20 cwt.	0·54				
	4. 2% foliage spray three times.	0·66				
	5. 2% foliage spray five times.	0·91				
	6. 1% foliage spray five times.	0·73				

SEASON 1946.

The following problems with regard to foliage spray treatments were further investigated. (1) Time of application and the minimum number of sprays required to be effective, (2) the possibility of incorporating magnesium sulphate in fungicides, such as Shirlan A.G. (anti-*Cladosporium* spray), when Mould is also a problem, (3) the rate of application and strength of spray required to give a satisfactory control, (4) avoidance of spray damage and the use of a variety of spreaders.

The soil dressings at 10 and 20 cwt. rates were also included during this season as standard applications to which the results of spray treatments could be referred.

The visual and chemical data are given in Table V and a statistical analysis of the chemical data is given in Table VI.

TIME OF APPLICATION AND THE MINIMUM NUMBER OF SPRAYS REQUIRED.

The data for centre A show that spraying at intervals of approximately three weeks, making a total of five sprays for the season, effected a complete control of the deficiency but that three early sprays were only effective in delaying the onset of symptoms. A spray applied when visual signs were first observed arrested their spread. Chemical values for the 20 cwt. application, the 2 per cent. spray three times, and the 2 per cent. spray five times, are significantly greater than those for the plants receiving no Mg at each date. The last named spray treatment gave significantly higher MgO values in the leaves than any other treatment. This is in accord with the visual data, and emphasizes the importance of a continued spray programme to counterbalance the deficiency completely.

At centre B, two early sprays prevented the development of visual symptoms until late season, and a spray at that stage checked the spread of symptoms, but a continuous spray programme was necessary to obtain complete success.

The order of significance at the first sampling date is 2 per cent. five times and 2 per cent. twice > 20 cwt. > control. Results for the 2 per cent. early spray treatment followed by a late one, suggest that the late spray increased the MgO content above the critical level, viz. 0·50 per cent., and that the 2 per cent. spray five times is significantly higher than any other treatment.

At centre C the superiority of a 2 per cent. foliage spray treatment, applied four times, is shown by the visual data and by the significantly higher MgO levels in the leaves, as compared with those from other treatments.

At centre D, three 2 per cent. sprays during early season delayed the onset of symptoms but it was necessary to apply a total of five sprays to effect a complete cure. Chemical data show that the MgO content of the leaves is in the following order of significance : 2 per cent. five times > 2 per cent. three times and 1 per cent. five times > 20 cwt. > 10 cwt. and untreated plots. The 1 per cent. five times and 2 per cent. three times have similar chemical values at the end of the season, and these are near the threshold of deficiency level, viz. 0·50 per cent MgO. The superiority of a 2 per cent. foliage spray applied five times, at intervals of three weeks during the growing season, is again shown at this centre.

INCLUSION OF MAGNESIUM SULPHATE IN FUNGICIDAL SPRAYS.

Although the supremacy of foliage sprays over soil treatment as regards both effectiveness and economy of material has been shown, commercial growers often prefer the soil treatment method because of the saving of time and labour during their busy season. To minimize the labour problem entailed in spraying, experiments were carried out in 1946 to test whether the inclusion of magnesium sulphate in the routine fungicidal spray, i.e. Shirlan A.G., was feasible. During the 1946 season duplicate Mg spray treatments were made at the four centres but in one block a fungicide, i.e. Shirlan A.G. (2 fluid oz. per 4 gallons) was also included in the sprays.

The visual and chemical data are given in Table V and the statistical analysis in Table VI. Spray treatments in Block A at the four centres included Shirlan A.G., whereas those of Block B had Mg only.

The data show that the Mg sprays were effective in the presence of the fungicide ; no spray damage was caused by the mixture. There are no significant differences in MgO content of the leaves between the two blocks at the four centres and this is confirmed by the visual symptoms recorded. This result may be important economically, especially when *Cladosporium* is also a problem, since the Mg spray including the fungicide corrected satisfactorily the deficiency and proved to be fungicidal.

RATE OF APPLICATION AND STRENGTH OF SPRAY REQUIRED.

The data obtained show that, as the plants grow, there is an increase in the volume of spray and in the quantity of salt used. The quantities of kieserite used in the sprays are relatively small as compared with those in the soil treatments, being usually less than 5 cwt. per acre for the season.

Two spray strengths have been tried at the four centres, viz. 1 and 2 per cent. During the winter of 1944 a 2½ spray was used at Centre D. The data in Tables III, IV and V show that the 2½ and 2 per cent. rates are equally effective and that both are superior to the 1 per cent. rate in the correction of the deficiency.

SPRAY DAMAGE AND USE OF SPREADER.

Spray damage occurred only when spraying was carried out on a dull day. In some instances, flower calyces were slightly injured but this had no adverse effect on the development of the affected fruitlets or on the yield of fruit. It is advisable to spray during sunny periods so that the spray dries quickly on leaf surfaces. As conditions may vary at different centres, it is advisable to apply trial sprays to plants a day before the main application to determine the likelihood of damage.

Ester salts (1 fluid oz. per 4 gallons) were used at centres A and B, and sulphonated lorol (1 oz. per 10 gallons) at centres C and D, during season 1946. The chemical data in Table V show that both are equally efficient as spreaders. It is essential to use a spreader so that the spray solution does not collect as droplets on the leaf surfaces otherwise leaf injury may occur. Microscopic studies showed that this injury resulted from a plasmolysis of the epidermal cells, especially the guard cells of the stomata which are the first to be affected.

DISCUSSION.

The main purpose of the experiments was to determine the most effective method of correcting magnesium deficiency in glasshouse tomatoes. The four centres chosen differed with regard to site, soil characters and manurial and cultural treatments but a soil analysis made at each centre showed that the exchangeable magnesium was low—0·009 to 0·014 per cent. MgO. This low magnesium status was corroborated by the presence of Mg deficiency symptoms in the tomato crops.

The methods used for the application of calcined kieserite (20 per cent. MgO) were: (1) applications of the solid to the soil at 5, 6, 10, 15 and 20 cwt. rates per acre; (2) 2 or 2½ per cent. solution applied to the soil, around the roots, at times and quantities as for foliage spray treatments; (3) foliage spray treatments with solutions of 1 and 2 per cent. applied either throughout the season, in the early season only, or later, after the onset of symptoms.

The results at the four centres showed that soil applications of magnesium sulphate at a minimum of 10 cwt. per acre of calcined kieserite (20 per cent. MgO) were necessary to effect a cure of commercial value. The 5 cwt. rate was unsatisfactory, whether applied in solution around the roots during the growth period or as a solid at planting time, the two methods of application giving similar results. There appeared to be little residual effect from a previous season's kieserite dressings, suggesting that there are soil factors which quickly immobilize the added Mg in the soil.

The superiority of foliage sprays over soil application, as regards both economy of material and efficiency, has been clearly demonstrated at all centres. The application of a solution of magnesium sulphate to the roots at times and in quantities as for foliage spraying failed to give such good correction of the deficiency.

The outstanding treatment was the 2 per cent. foliage spray, applied five times at intervals of approximately two to three weeks during the season, starting at the early growth stage. In many instances, using as little as 3 cwt. per acre of material, this treatment completely overcame the deficiency. This shows that large proportions of the heavy soil dressings of kieserite are unavailable to the plants.

It is clear that early sprays can delay the onset of symptoms and that a spray applied when symptoms first appear can arrest their development, but in severe cases a number of sprayings is required to give a full cure. A 2 per cent. spray is more effective than a 1 per cent. when both are applied five times during the season, whereas a spray strength greater than 2 per cent. is liable to cause damage.

The results of the spray experiments including a fungicide (Shirlan A.G.), show that the spray mixture, which includes additional spreader, corrects the deficiency without foliage damage and at the same time is fungicidal. This result is important in considering the relative economic merits of soil dressings versus foliage spray treatments when Mould is also a problem. Measures for correcting mineral deficiencies should, as far as possible, be linked with routine commercial practices and future work should consider this important aspect. Spray injury occurred when spraying was done during dull periods, apparently due to the persistence of spray droplets on leaf surfaces causing plasmolysis of the epidermal cells.

The data presented here may be compared with those obtained by other workers. Cromwell and Hunter (1) have reported that Mg deficiency in tomato is widespread in the west of Scotland and suggest that the deficiency in the leaves is a result of a physiological drain of the available magnesium from mature to immature leaves and fruit. They also suggest that K dressings accentuated the deficiencies and showed that the replaceable Mg was ample in the soil in which chlorotic plants were growing. In the present experiments, however, the depressing effect of K on Mg, as judged by leaf analysis and visual data, was not a crucial factor but the exchangeable Mg in the soil was initially low. Walsh and Clarke (13, 14) have also directed attention to the importance of K/Mg ratio in tomato problems. The present results show that in dealing with the deficiency the most important point is to provide an adequate supply of magnesium.

Hunter (2) has recently reported that heavy dressings of magnesium sulphate to tomato soils have been ineffective in Scotland and that the spray method would be unlikely to be of practical value in that area. Cultural treatments, such as early mulching with farmyard manure and peat, together with resoiling and reducing potash levels, are suggested as possible measures to offset Mg deficiency. On the other hand, the efficiency and economy of spray treatments as herein reported appear to make them preferable.

Owen (10) states that there are several types of Mg deficiency symptoms in tomato: (1) occurring early and in mid-season, associated with the physiological strain, (2) a more permanent type occurring later in the season due to undesirable physical conditions in the subsoil, (3) occurring at all stages. It is also stated that types (1) and (2) may be cured by adding nitrogenous fertilizers. A similar effect has been noted in the PK (omit N) plots at Long Ashton where susceptible crops, e.g. potato, cauliflower and apple readily show Mg deficiency symptoms in the absence of nitrogen. Chemical analysis and visual data show that the symptoms are due to Mg deficiency and that N applications increase the Mg content of the plants (6, 9).

The experiments also show the value of using the visual and chemical methods in complementary roles in assessing the results of the different treatments. The rapid tissue test (5, 6, 9) has proved invaluable as a quick method to supplement the visual data, and full chemical analyses have confirmed closely the results of the two methods. From the chemical data it seems possible to suggest a value of 0·50 per cent. MgO as an approximate threshold level of deficiency for the mid-stem leaves, as values below this were usually associated with the development of visual symptoms. This value, used as a rough dividing line between deficiency and sufficiency, was applicable to the data for the four years, irrespective of centre and variety used.

SUMMARY.

1. Experiments on the control of magnesium deficiency in glasshouse tomatoes were made at four centres, differing in cultural and manurial treatments and including a number of tomato varieties.

2. A minimum of 10 cwt. per acre of calcined kieserite (20 per cent. MgO) applied to the soil was necessary to achieve a commercially valuable correction of the deficiency and in instances of serious deficiency 20 cwt. per acre was more satisfactory.

3. The application of magnesium sulphate in solution to the roots, at times and amounts as for foliage spray treatments, proved less effective than the latter, and was similar in effect to applications of the solid to the soil at the same rate, at planting time.

4. The most effective treatment was a 2 per cent. spray applied to the foliage five times at intervals of two to three weeks, commencing at an early stage of growth. The total quantity of magnesium sulphate used during the season in this treatment

was less than 5 cwt. per acre. This suggests that soil factors immobilize a large proportion of the soil dressings of $MgSO_4$.

5. Early sprays were useful in delaying the onset of symptoms and late sprays in arresting their development.

6. A 2 per cent. concentration was more effective than a 1 per cent. as a foliage spray but a rate above 2 per cent. may cause leaf damage.

7. Risk of spray injury is decreased by spraying on sunny days when quick drying occurs. Trial sprayings are recommended before the main applications.

8. Plasmolysis of the epidermal cells, especially the stomatal guard cells, occurred when spray droplets remained on the leaf surfaces. Ester salts and sulphonated lorol were equally efficient as spreaders.

9. A magnesium sulphate spray plus a fungicide, Shirlan A.G. (4 fluid oz. per 4 gallons), proved as effective as the magnesium salt used alone in the correction of the deficiency, and the fungicidal value of the Shirlan was not decreased.

10. The incidence of Mg deficiency is reduced by soil application of calcined kieserite with or without potash. The Mg content of the leaves is slightly higher when K is omitted but the omission of K only does not cure the deficiency.

11. The reported correction of Mg deficiency, using nitrogenous fertilizers, is probably an indirect effect. Symptoms of Mg deficiency occur in susceptible crops under PK (omit N) treatments at Long Ashton but not in those receiving N. Sand culture experiments also show that Mg intake has been decreased by withholding N supply.

12. The value of the visual and chemical methods in these experiments is shown. Chemical data on the mid-stem leaves suggest that 0·5 per cent. MgO may be regarded as a threshold of deficiency level characterized by the development of visual symptoms.

ACKNOWLEDGMENTS.

The work was carried out with the aid of Special Grants from the Agricultural Research Council to whom grateful acknowledgment is made. Professor T. Wallace was responsible for the general direction of the work and the author's thanks are due to him for valuable advice during the course of the experiments. Mr. J. O. Jones, now Advisory Chemist, South Eastern Province of the N.A.A.S., collaborated with the author from 1943 to 1945 inclusive. The assistance given by the growers at the four centres is gratefully acknowledged.

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(*Received* 26/1/48.)

A METHOD FOR THE QUANTITATIVE ESTIMATION OF GROWTH SUBSTANCES BASED ON THE RESPONSE OF TOMATO OVARIES TO KNOWN AMOUNTS OF 2-NAPHTHOXY-ACETIC ACID

By **L. C. LUCKWILL**
Long Ashton Research Station, Bristol

SYNOPSIS.

Unpollinated tomato ovaries treated with certain synthetic growth substances can be stimulated to develop into seedless fruits, and the rate of development during the first six days is shown to be related to the amount of growth substance with which the ovary is treated. The details of this relationship have been worked out in a series of experiments in which ovaries were treated with known amounts of 2-naphthoxy-acetic acid (BNOA). With this as a basis, a quantitative test method has been evolved by which the activity of different growth substances and natural hormone extracts can be measured relative to that of BNOA. The method, as is described in the next article in this number (see p. 32), has been used to follow the changes in the hormone content of apple seeds throughout the season.

INTRODUCTION.

In the study of induced parthenocarpy the tomato has been widely used as a test object, since it is a plant in which pollination can be easily prevented and one in which parthenocarpic development of the fruit is readily stimulated by a wide range of synthetic growth substances.

In work of this type the growth substance is usually applied to the ovaries of emasculated flowers, either as a fine spray or aerosol, or in the form of a lanolin paste or emulsion, and the minimum concentration required to induce the development of fruits (the " threshold " concentration) is taken as a measure of the activity of the substance being investigated. Although useful as purely qualitative methods, these techniques cannot be regarded as satisfactory for quantitative work. Their chief disadvantage lies in the fact that it is not possible to treat each ovary with a standard amount of growth substance or to measure the quantity of paste, emulsion or spray which has been applied. Moreover, it is likely that the responses of plants of different ages, varieties, and states of health will be different, and, unless the conditions of growth can be carefully controlled, the results of experiments carried out at different times and by different workers will not be comparable. Finally, there is the serious disadvantage of having to wait until the fruits are fully grown, a period of forty to fifty days, before the activity of the substance under test can finally be assessed.

It was in order to overcome difficulties such as these and to place the " tomato ovary test " on a more strictly quantitative basis, that the work described below was carried out. It was eventually found possible to reduce the time required for carrying out a test to six days, and to obtain comparable results from experiments carried out at different times in ordinary greenhouses without temperature,- light- or humidity-control. The method is at present in use for obtaining data on the relative fructigenic activity of various substituted naphthoxy- acids, and for a study of a natural hormone extracted from developing apple seeds (3), and has been found to give very satisfactory results.

BASIS OF THE METHOD.

The method of estimation here described depends on the fact that the rate of growth of an unpollinated tomato ovary treated with growth substance is related to the quantity of growth substance applied : so that by finding the amounts of different growth substances required to produce the same rate of growth, it is possible to measure their relative activities. A method is described whereby variations in growth rate in different experiments, due to different environmental conditions, can be taken into account, and figures are given for the construction of dosage/response curves for 2-naphthoxy-acetic acid (BNOA). It is suggested that this substance, because of its specific action for parthenocarpy, should be taken as a standard with which the fructigenic activity of other growth substances can be compared.

METHOD.

Material. In order to ensure the greatest possible uniformity of material for use in these tests, a strain of tomato derived from the commercial variety " Blaby ", which had been inbred for many generations, was used. This is referred to as Line 107. For some of the preliminary work plants of the variety " Potentate " were used and a few trials were also carried out using an inbred line of the Red Currant Tomato (*Lycopersicon pimpinellifolium*).

Culture of plants. Seeds were sown in boxes, pricked off in the cotyledon stage into 4-inch pots and later transplanted into 7-inch pots for flowering. John Innes seed- and potting-composts were used throughout. Only flowers on the first and second trusses were used in these experiments, and growth was stopped two leaves beyond the second truss : by this means it was possible to avoid the need for staking and for further transplantation into larger pots, which would have meant reducing the scale of the experiments. Side shoots were kept pinched out and, during the summer months, as soon as the first truss appeared, the plants received weekly applications of dried blood to keep them in vigorous condition. Usually about 30 per cent. more plants than were required for each test were raised, which enabled the uniformity of the material to be further increased by discarding the most and the least advanced plants in each batch.

Lay-out of experiments. As a precaution against possible variations in temperature in different parts of the greenhouse the experiments were always laid out in randomized blocks with each treatment replicated two to eight times in each block. The results were examined using the technique of analysis of variance, but in none of the experiments were significant block variances found.

Preparation of the plants for test. As soon as the oldest flowers on the first truss started to open the plants were prepared for use. Two flower buds at a stage of development one to two days before flower opening were chosen and were emasculated by removing the stamen tube, together with the corolla and the style, with the fingers. All other flowers and buds on the truss were then cut off. In selecting ovaries those which were fasciated or of irregular shape were discarded, and in this manner, it was possible to obtain a series of ovaries of great uniformity, both as regards size, shape, and stage of development. The ovaries were treated with growth substance within twenty-four hours of preparation.

Treating ovaries with a known quantity of growth substance. The growth substances were applied to the ovaries in the form of aqueous solutions, without the addition of wetting agents, by means of a small pipette of standard aperture. A 1 ml. hypodermic syringe fitted with a number 17 needle was found to be very satisfactory for this purpose, as it delivered drops of constant size (0·0075 ml.) and the rate of dropping could be readily controlled by manipulating the plunger (Plate I). During treatment the flower was held in a vertical position and the required number of drops allowed to fall on to the ovary, where they remained held by surface tension after the flower returned to its normal inverted position. The standard procedure adopted in carrying out the tests was to treat each ovary with three drops (0·0225 ml.) of solution containing the required amount of growth substance.

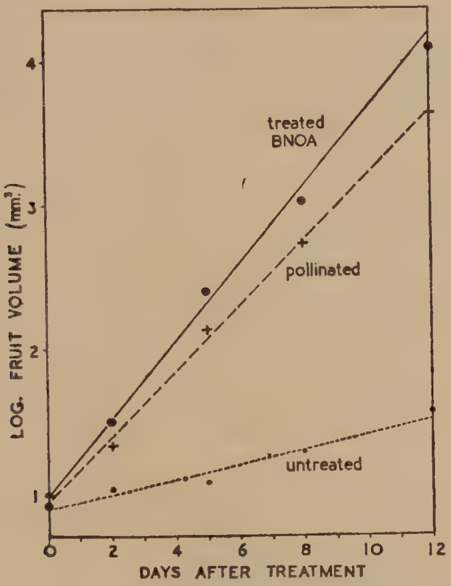


FIG. 1.

Logarithmic growth curves of tomato ovaries during the first twelve days. The growth of both treated and untreated ovaries is exponential. Each curve is based on the mean of six ovaries.

Mode of growth of the fruit. Preliminary studies showed that the growth of the tomato ovary following pollination or treatment with growth substance is strictly exponential up till about the 12th day (Fig. 1), after which the relative growth rate gradually falls. Ovaries which are neither pollinated nor treated with growth substance also grow exponentially, but at a very much slower rate. The early growth of the fruit can therefore be expressed by the general formula:

$$r = \frac{\log (V/v)}{t}$$

where r=relative growth rate; v=volume of ovary before treatment; and V=volume of fruit after t days.

Since the ovaries and young fruits are regular in shape and approximately spherical, little error will be introduced by regarding the volume as a constant function of the cube of the diameter (d), in which case the growth equation can be written :

$$r = \frac{\log (kD^3/kd^3)}{t} = \frac{3 \log (D/d)}{t}$$

Throughout this investigation t was kept constant at six days, so that the working equation could be still further simplified to :

$$r = \tfrac{1}{2} \log (D/d).$$

Measurement of response. The response of ovaries to varying doses of growth substance was measured as the increase in the relative growth rate of the treated ovaries, over that of a control series treated with water only. Thus, if r_x=mean r for treated ovaries and r_c=mean r for controls, then :

$$\text{Response} = r_x - r_c = \tfrac{1}{2} \log (D_x/d) - \tfrac{1}{2} \log (D_c/d).$$

Over a series of experiments it was found that d, the initial diameter of the untreated ovaries, varied so little that, for practical purposes it could be regarded as constant, so that :

$$\text{Response} = h \propto \log D_x - \log D_c.$$

As a result of this simplification the only measurements required for estimating the response of ovaries to different amounts of growth substance are the mean diameters of the treated and of the control ovaries after six days' growth.

In comparing the results of experiments carried out at different times, variations in the growth rate of the ovaries due to different environmental factors, particularly temperature, were eliminated by expressing the observed response h as a percentage of H, the maximum response for each particular test. The latter was determined graphically by extrapolation of the curve showing variation of h with the logarithm of the dose (see Fig. 4).

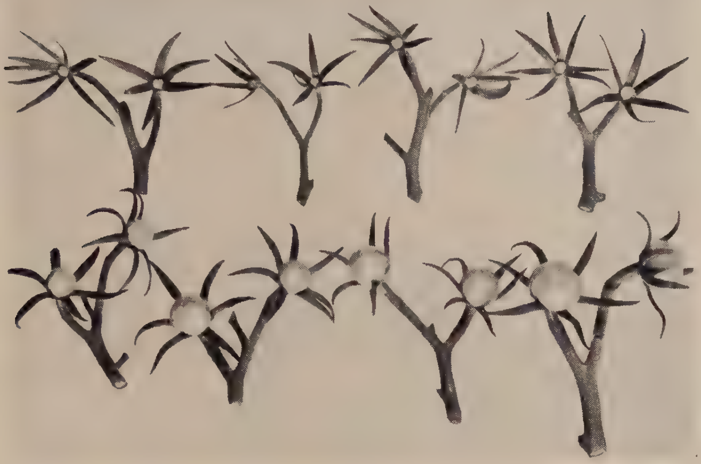
In carrying out the tests the ovaries were cut from the plant six days after treatment and their diameters were measured under a binocular dissecting microscope fitted with an eyepiece scale, at a magnification of × 8. A typical series of ovaries, six days after treating with 2-naphthoxy-acetic acid in doses ranging from 0·0112 to 2·25 μg. per ovary is shown in Plate I.

EXPERIMENTAL DATA.

Concentration and dose. The fact that tomato ovaries treated with the same volume of growth substance solution of different concentrations display varying rates of growth may mean that the growth rate is influenced either by the concentration of the growth substance in the solution, or by the total quantity of growth substance with which the ovary has been treated (i.e. the dose), or possibly by both of these factors. In order to decide between these possibilities preliminary experiments were carried out in which either the concentration or the dose was varied, the other factor being held constant.

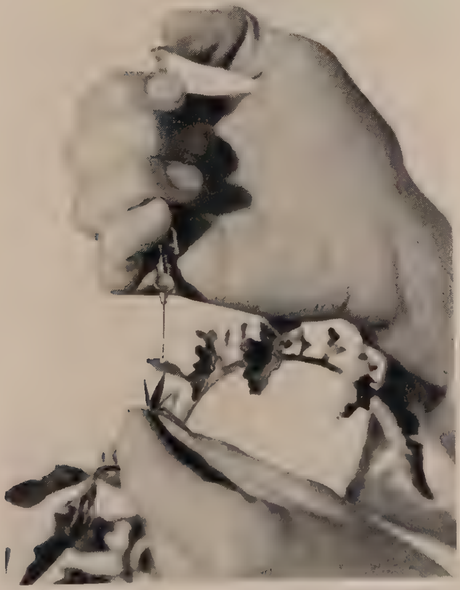
Variations in dose at constant concentration were achieved by treating ovaries with different quantities (from one to six drops) of BNOA solution of strength 20 p.p.m., giving doses ranging from 0·15 to 0·90 μg. In the second experiment the dose was kept constant at 1·0 μg. BNOA per ovary and the concentration varied by adding different volumes of solutions ranging from 22·2 to 133·3 p.p.m. Each dose and concentration was tested on twenty ovaries.

PLATE I.



(*Above.*) A typical series of tomato ovaries photographed six days after being treated with the following doses of 2-naphthoxy-acetic acid (*starting from top left*): 0, 0·0112, 0·0225, 0·1120, 0·2250, 0·560, 1·125, 2·250 μg. per ovary.

(*Below.*) Method of using hypodermic syringe for treating ovaries with known amounts of growth substance.



The results of these two experiments are given in graphical form in Figs 2 and 3, and show quite clearly that the growth rate of the young fruit depends on the *quantity* of growth substance applied (i.e. the dose) and not directly on the concentration of the solution used. Although there appears from Fig. 3 to be a slight increase in growth rate with increasing log. concentration, statistical tests show that this is of no significance.

It should be noted that in all subsequent tests the dose was varied by using solutions of different concentrations and keeping the volume of the solution constant at three drops (=0·0225 ml.) per ovary.

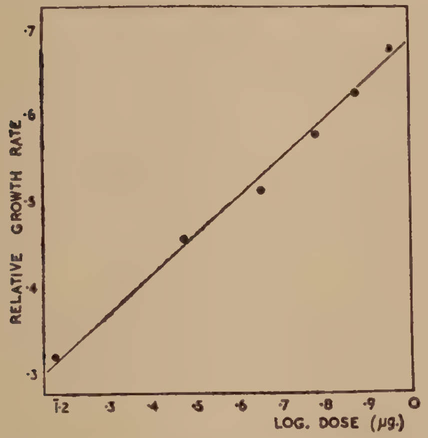


Fig. 2.

Relationship between the relative growth rate of the ovary and the dose of BNOA administered. The concentration of the solution was kept constant. Each point is based on the mean of twenty ovaries.

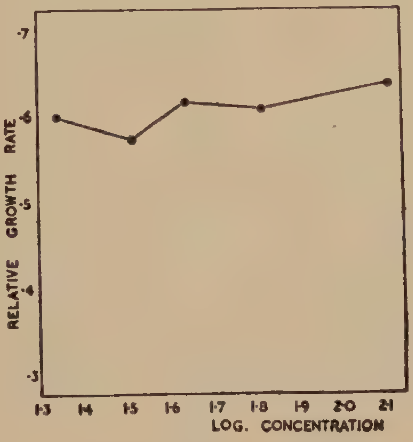


Fig. 3.

Relationship between the relative growth rate of the ovary and the concentration of BNOA in the solution. The dose was kept constant. Each point is based on the mean of twenty ovaries.

Relation between dose and response. It will be noted from Fig. 2 that the growth rate of the young fruit is proportional to the logarithm of the dose of BNOA administered. This linear relationship, however, was found to hold only over a limited range of dosage, and, over a wider range, the relationship always assumed the sigmoid form of a typical dosage/response curve.

The results of a series of eight experiments carried out with BNOA at different times of the year are presented in Table I, and the data for one of these experiments (No. 4) is shown graphically in Fig. 4. Using the standard methods of probit analysis, modified for quantitative data (2), a normal sigmoid curve has been fitted to the points and it will be seen that the agreement between the observed and expected responses is good. In some experiments, however, the observed responses to very low doses tended to be higher than those expected on the assumption of normal distribution (see Table I). In order to test the significance of these deviations, a " heterogeneity variance ", based on the deviations of the observed from the expected responses, was calculated, and compared with an " error variance " derived from variations of log. diameters " within doses ", by means of the variance ratio test. The results showed that in only one experiment (No. 5) did the deviation of the

TABLE I.

Observed responses (h) as percentages of the maximum response (H) of tomato ovaries treated with varying doses of BNOA, compared with responses expected on the assumption of a normal sigmoid log. dose/response relationship.

Exp. No.	Date.	No. of ovaries per treatment.		Dose (μg. per ovary).									
				0·0112	0·0225	0·056	0·112	0·225	0·560	1·125	2·25	5·60	H.
1	2/5	20	Obs. %	2·5	3·0	—	13·2	42·5	—	98·8	98·6	—	0·528
			Exp. %	0·0	0·0	—	13·6	42·5	—	97·3	99·8	—	
2	31/5	8	Obs. %	—	4·0	—	34·1	54·7	—	94·9	99·8	—	0·756
			Exp. %	—	3·4	—	33·4	56·8	—	94·0	98·5	—	
3	23/7	16	Obs. %	3·9	3·5	12·4	23·1	48·5	92·0	94·0	99·3	98·7	0·710
			Exp. %	0·1	0·8	7·8	24·8	52·8	85·6	96·4	99·5	—	
4	21/8	32	Obs. %	—	2·2	2·2	12·1	59·4	89·5	97·0	99·8	—	0·685
			Exp. %	—	0·1	3·4	18·7	51·6	90·1	98·7	99·9	—	
5	29/8	32	Obs. %	—	3·7	12·8	18·0	28·8	75·0	88·8	97·5	—	0·510
			Exp. %	—	0·5	4·7	15·6	38·1	71·6	91·3	97·3	—	
6	11/9	28	Obs. %	3·7	4·9	9·7	15·7	32·1	75·3	92·8	99·3	96·0	0·465
			Exp. %	0·1	0·4	4·1	15·2	37·1	73·2	90·7	97·9	—	
7	25/9	16	Obs. %	—	—	8·9	22·4	31·4	65·3	91·7	96·5	78·0	0·318
			Exp. %	—	—	5·8	16·6	36·0	67·4	85·6	95·2	—	
8	12/11	16	Obs. %	2·6	10·7	14·1	14·1	28·2	53·8	74·8	90·2	97·6	0·234
			Exp. %	0·4	1·6	6·8	15·9	27·1	56·0	73·6	86·9	96·2	

observed from the expected responses exceed the 5 per cent. level of significance; in this particular case the bulk of the heterogeneity variance could be attributed to the exceptionally wide deviation of a single point of the curve (viz. that corresponding to a dose of 0·056 μg).

It may therefore be concluded that the normal sigmoid curve is a satisfactory representation of the relationship between the logarithm of the dose of growth substance administered and the response of the ovary, at least for doses up to 2·5 μg. BNOA. At still higher doses there is a tendency for the growth rate to decrease again (see Table I) but investigation of the trend of the curve in this direction is limited by the solubility of BNOA in water.

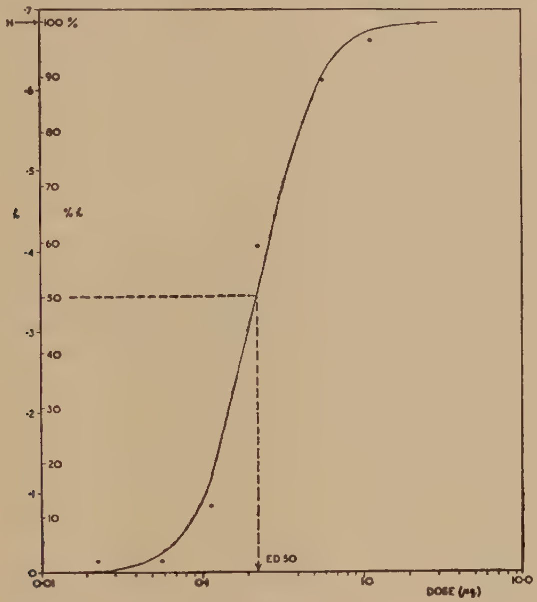


FIG. 4.

The relationship between the dose of BNOA administered and the response of the ovary in a typical experiment (No. 4). A normal sigmoid curve has been fitted to the points.

The median effective dose. Following the standard terminology used in toxicology, where similar dosage/response relationships hold, the median effective dose (ED 50) may be defined as that dose of growth substance which will induce a response equal to half the maximum response under the particular conditions of the experiment (see Fig. 4).

The ED 50 for BNOA as determined from different experiments was found to vary over wide limits (Table II), and statistical tests showed that these variations were highly significant. It was further observed that the ED 50 showed a strong negative correlation with the maximum growth rate (H) as estimated by extrapolation of the sigmoid curve. When log. ED 50 is plotted against H (Fig. 5) the regression is seen to be linear. The following statistics were calculated:

Correlation coefficient: log. ED 50/H = − 0·971.
Regression coefficient: log. ED 50 on H = − 0·6872.

TABLE II.

Values of the probit regression coefficient and median effective dose for BNOA in eight different experiments.

Experiment number.	Date.	H.	Slope of probit regression line.		Median effective dose.	
			b	S.E.	ED 50	5% fiducial limits.
2	31/5	0·756	1·99	0·57	0·185	0·132–0·260
3	32/6	0·710	2·47	0·16	0·210	0·192–0·230
4	21/8	0·685	3·02	0·21	0·216	0·198–0·235
1	2/5	0·528	3·03	0·36	0·258	0·224–0·297
5	29/8	0·510	2·25	0·14	0·312	0·284–0·342
6	11/9	0·465	2·35	0·23	0·307	0·265–0·357
7	25/9	0·318	2·02	0·39	0·336	0·250–0·451
8	12/11	0·234	1·63	0·21	0·458	0·362–0·579
	Overall mean*	—	2·29	0·32		

* Calculated from the variances and co-variances for each experiment.

It will be seen from Table II that in November, when the growth rate of the fruit was very low, approximately 2·5 times as much BNOA was required per ovary to elicit the same percentage response as in early summer, when the conditions for growth were optimum. This observation is in line with general experience in the practical application of growth substances for weed-killing and for the control of pre-harvest drop of fruit, where it is found that the best response is obtained when the temperature at the time of application is high and growth is vigorous, and that where the growth substances have to be applied in cool weather it is necessary to use higher concentrations to get the desired effect.

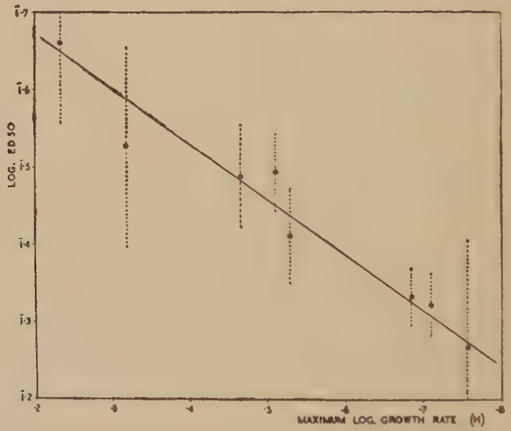


FIG. 5.
Regression of log. ED 50 on H, the maximum growth rate. The dotted lines represent a range of ±2 × S.E.

The values of b, calculated from different experiments, show a rather wide range of variation, but in no case does the difference between b and the mean b exceed twice the standard error of the difference. The variations cannot therefore be considered statistically significant.

Construction of standard dosage/response curves for BNOA. From the above results it will be appreciated that, for the complete description of the dosage/response curve for any given growth substance, two statistics must be known, viz. :

(1) b, the slope of the probit regression line, which defines the form and slope of the curve. For any given substance this appears to be constant, within the limits of statistical variation, under a wide range of environmental conditions, and for BNOA the mean value of b=2·29 ± 0·32 may be taken as characteristic. The value for b for other growth substances may, of course, differ from that for BNOA.

(2) The ED 50. This determines the position of the curve on the abscissa and will vary from experiment to experiment. The ED 50 for BNOA can be estimated from H, the maximum response, with the aid of the regression diagram shown in Fig. 5, or, more conveniently from Table III.

TABLE III.

Relation between the maximum response (H) and the median effective dose (ED 50) for 2-naphthoxy-acetic acid.

Maximum response. H.	ED 50 μg. per ovary.
0·2	0·460
0·3	0·393
0·4	0·336
0·5	0·286
0·6	0·244
0·7	0·209
0·8	0·178
0·9	0·152

For routine estimations little error will be introduced by taking H as the mean response of ovaries treated with 2·25 μg. BNOA (=0·0225 ml. × 100 p.p.m.), as during the period from May till the middle of September the mean response to this dose varied between 98·6 and 99·8 per cent. of the maximum response as determined graphically (Table I). For experiments at other times of the year it is recommended that the BNOA controls should receive a dose of 5·6 μg. per ovary.

PROCEDURE FOR ESTIMATING ACTIVITY OF A GROWTH SUBSTANCE.

The procedure for determining the activity of a growth substance relative to that of BNOA may now be summarized :

(1) Preliminary qualitative tests are carried out to determine the approximate range of concentrations over which the substance shows activity.

(2) A suitable series of five or six dilutions are made up, and 0·0225 ml. is placed on each of the test ovaries. Each dilution is tested on at least ten ovaries.

(3) Control series of ovaries are treated with (*a*) 0·0225 ml. water, and (*b*) 0·0225 ml. 2-naphthoxy-acetic acid 100 p.p.m. (=2·25 μg).

(4) After six days the diameters of the young fruits are measured and, from the mean log. diameters, the percentage response for each dilution is calculated and plotted against the logarithm of the dose.

(5) Using the probit technique a normal sigmoid curve is fitted to the points and from this curve (or from the probit regression line itself) the ED 50 is read off.

(6) The response of the ovaries treated with 2·25 µg. BNOA is taken as H, and from this the corresponding ED 50 for BNOA can be estimated from Table III or from a graph.

(7) The ratio of the reciprocals of the ED 50's for the two different substances will give a measure of their relative potencies.

The results of a typical experiment in which the activity of two growth substances, 2:4 dichlorophenoxy-acetic acid and α-(2-naphthoxy-) propionic acid have been compared with that of 2-naphthoxy-acetic acid are shown in Fig. 6. The relative potencies are as follows:

Substance.	ED 50.	1/ED 50.	Relative potency.
2:4 dichlorophenoxy-acetic acid	0·018	55·5	1030
2-naphthoxy-acetic acid	0·186	5·4	100
α-(2-naphthoxy-) propionic acid	2·000	0·5	9

It should be noted that where, as in the present instance, the dosage/response curves for the different substances are not parallel, the relative potencies will depend on the level of response at which the estimation is made. The relative potencies estimated from the ED 75 or ED 25, for example, will not necessarily be the same as those estimated from the ED 50. In comparing the activity of different substances, therefore, it is necessary to bear this in mind and to state the response level at which the estimations have been made.

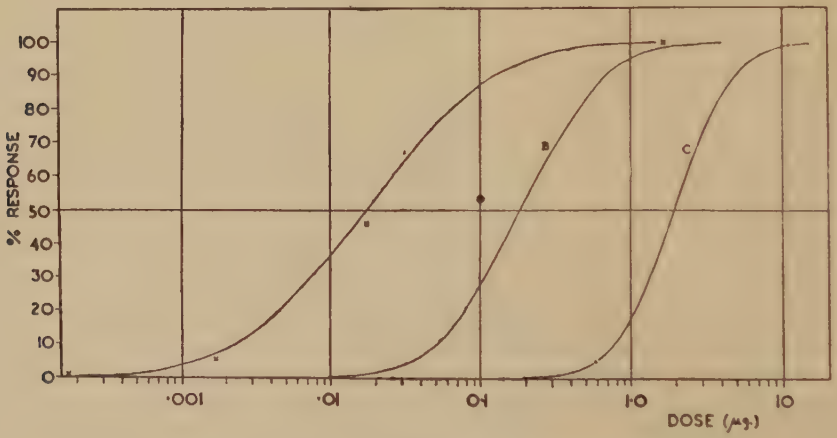


FIG. 6.

Dosage/response curves for 2:4 dichlorophenoxy-acetic acid (A) and α-(2-naphthoxy-) propionic acid (C) compared with the mean curve for 2-naphthoxy-acetic acid (B).

Estimation of hormone content of apple seeds. In estimating the hormone content of apple seeds (3) an extract of 500 seeds was concentrated to 1 ml. and from this solution a series of dilutions corresponding to 167, 50, 17 and 5 seeds per ml. was made. The percentage response for each solution was found and plotted against the log. concentration (expressed as seeds per ml.). The result was a sigmoid curve, the slope of which was found not to differ significantly from that of the standard BNOA dosage/response curve (Fig. 7). From this curve the 50 per cent. concentration was read off and, by comparing this with the corresponding ED 50, estimated from the BNOA control, it was possible to express the hormone content of the seed as μg. equivalents of BNOA. Thus, for the example illustrated in Fig. 7:

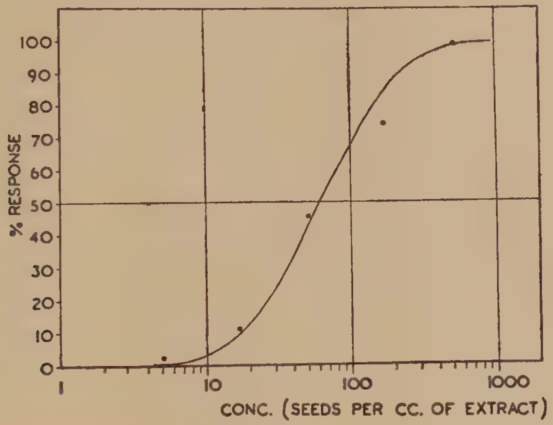


FIG. 7.

Results of an experiment with an extract prepared from the seeds of the apple variety Miller's Seedling. The curve is the mean dosage/response curve for BNOA, which is seen to fit the experimental points closely.

50 per cent. concentration =60 seeds per ml.
=60 × 0·0225 seeds per ovary.
=1·35 seeds per ovary.

ED 50 for BNOA =0·246 μg. per ovary.

Hence 100 seeds contain the equivalent of $\frac{100}{1\cdot35} \times 0\cdot246$ μg. BNOA.
=18·2 μg. BNOA.

DISCUSSION.

A number of methods for the biological estimation of growth substances are already available, amongst which may be mentioned the standard Avena test (6), the Avena coleoptile cylinder test (1), the Pea test (7), and the Corn root test (4). All of these, however, are based on the ability of the substance to stimulate or inhibit cell extension and are therefore unsuitable for measuring fructigenic activity, i.e. activity in stimulating parthenocarpy. It is for this reason that the method outlined above was devised. Although some substances such as 2:4 dichlorophenoxy-acetic acid, are active both for cell extension and for parthenocarpy, others, such as 2-naphthoxy-acetic acid are specific for parthenocarpy. Indolyl-acetic acid, on the other hand, which is one of the most active compounds in the standard Avena test, was found to be relatively inactive on tomato ovaries.

In any method for the biological estimation of growth substances two factors must be taken into account, viz. :

(1) The primary activity of the growth substance.

(2) The rate of transport of the growth substance between the point of application and the seat of action.

Where information on the primary activity of a series of growth substances is required it is clearly necessary to use a test in which any effects due to differential rates of transport are reduced to a minimum. As pointed out by Veldstra (5), the Pea test, in which the test material is immersed in a solution of the growth substance, fulfils this requirement, whereas the standard Avena test does not. The method here described also seems to be satisfactory in this respect, and the fact that the concentration of the solution is without direct influence on the response of the ovary confirms that transport of the growth substance into the fruit is not a limiting factor in this test. The method has the further advantage that it can be carried out in ordinary glasshouses without the necessity for temperature- and humidity-control, though no doubt where such control is available the precision of the test could be still further increased.

SUMMARY.

1. The essential features of the method described are :

(i) Treatment of unpollinated tomato ovaries with known amounts of growth substance in aqueous solution.

(ii) Measurement of the increase in growth rate of the treated ovaries over that of the controls, during a period of six days.

2. The increase in the growth rate of the ovary is related to the dose of growth substance administered, but does not depend directly on the concentration of the solution used.

3. The relationship between the response of the ovary and the logarithm of the dose can be represented by a normal sigmoid curve.

4. For any given growth substance the slope of the dosage/response curve is constant, but its position on the abscissa, as measured by the median effective dose (ED 50), varies from experiment to experiment.

5. Over a series of eight experiments with 2-naphthoxy-acetic acid the ED 50 showed a strong negative correlation ($r = -0{\cdot}971$) with the maximum growth rate, varying from 0·458 μg. per ovary in November to 0·185 μg. in May when conditions for growth were optimum.

6. Examples are given showing how these data can be used for estimating the relative fructigenic activity of different growth substances, and for measuring the natural hormone content of apple seed extracts.

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(*Received* 26/2/48.)

THE HORMONE CONTENT OF THE SEED IN RELATION TO ENDOSPERM DEVELOPMENT AND FRUIT DROP IN THE APPLE

By **L. C. LUCKWILL**
Long Ashton Research Station, Bristol

SYNOPSIS.

Apple seeds at certain stages of development have been shown to contain a hormone which can be extracted with boiling water and which is active in stimulating the development of unpollinated tomato ovaries. Use has been made of this latter fact to detect and measure the amount of hormone present in the seeds at various stages of development, using the methods described in the previous paper (see pp. 19 to 31).

In the variety Beauty of Bath the appearance of the hormone in the seed at 25-30 days after petal-fall corresponded with the cessation of the post-blossom drop, and the disappearance of hormone from the seeds of nearly ripe apples was correlated with the occurrence of the pre-harvest drop. The seeds from " June drop " fruitlets of two other varieties contained much less hormone than those from fruitlets remaining attached to the tree, and from this and other evidence it is concluded that this hormone is concerned with the control of fruit drop. It appears to be produced in the endosperm tissue of the seed but not in the embryo. Although it will stimulate the development of tomato ovaries it apparently plays no direct part in initiating fruit growth in the apple. The presence of a second, as yet undetected, hormone with this function is postulated.

INTRODUCTION.

The discovery that it is possible to extract from apple seeds at certain stages of development a hormone-like substance which is active in stimulating parthenocarpy in the tomato was announced in a short note published in 1946 (5). The present paper sets forth the results of further work carried out during 1947 with the apple variety Beauty of Bath, in which an attempt was made to correlate the changes in the hormone content of the seed throughout the season with seed and fruit growth, and with fruit drop. This work was made possible by the development of a quantitative assay method, which has been described in a previous paper (6) and which was based on the rate of development of tomato ovaries when treated with known amounts of growth substance.

EXPERIMENTAL METHODS.

PREPARATION OF SEED EXTRACTS.

The seeds were extracted from representative samples of lateral fruits immediately after picking. Within ten minutes of removal from the fruit, before appreciable browning had set in, the seeds were dried in an oven at 110-120° C. After drying they were ground to a fine powder and stored in corked tubes until required for extraction.

Extraction of the seed samples was carried out as soon as possible and never more than fourteen days after drying. A weight of powdered seeds corresponding to 500 seeds, or, in the earlier stages, to 1,000 seeds, was boiled with six to ten times its weight of water in an open beaker for fifteen minutes. Whilst still hot the extract was filtered off through glass wool and the residue reboiled with a further volume of water for another fifteen minutes. The water extracts were bulked and were then shaken four times with ten times their volume of peroxide-free ether. The ether layers were separated, bulked, and distilled to dryness in a tared 2 by $\frac{5}{8}$ in. specimen tube held in a water bath at 70° C.

The dried ether extract was redissolved in exactly 1 ml. distilled water at 70° C. and the solution cooled to just above freezing point. On cooling, a dense precipitate consisting principally of the glucoside phloridzin (10) was obtained and was removed by filtration. The filtrate, containing the active principle, was stored at 5° C. until required for testing.

METHOD OF TESTING EXTRACTS.

The extracts prepared as above contained the active principle from 500 seeds concentrated in 1 ml. of fluid. From this original extract a series of dilutions were made up representing concentrations of 167, 50, 17 and 5 seeds per ml. The original extract and each of the four dilutions were tested on a series of eight or ten tomato ovaries, a quantity of 0·0225 ml. of liquid being placed on each ovary. Control series of ovaries were treated with water and 2-naphthoxy-acetic acid (BNOA) 100 p.p.m., and the tests were carried out using the standard procedure described elsewhere (6). The hormone content of the seeds was expressed as μg. equivalents of BNOA per 100 seeds, and the concentration of hormone in the seeds as μg. equivalents of BNOA per gm. dry weight.

MATERIALS FOR MORPHOLOGICAL WORK.

At each sampling about one dozen seeds taken at random from the sample were fixed in Flemming's chrom-osmium-acetic fixative (medium strength). These were embedded in paraffin wax, sectioned at 14 μ and stained in Delafield's haematoxylin to provide material for the study of seed development.

At the same time a number of representative fruits from each sample were preserved in 70 per cent. alcohol for the study of fruit development. Later, vertical sections of the fruit were cut several mm. thick and were cleared in lacto-phenol to show up the internal structures.

The exceptionally short blossoming period in 1947 resulted in an unusually low variability of the fruit and seed samples and materially assisted the investigation.

MEASUREMENT OF FRUIT-DROP.

Representative branches of five different trees were tagged at full-bloom and a count was made of the total number of blossoms on these branches. Counts of the number of fruits remaining on these branches were made at approximately weekly intervals throughout the season.

RESULTS.

HORMONE CONTENT OF SEED.

The trees blossomed during the second week in May and the average date of petal fall was May 17th. On August 15th, i.e. ninety days later, the fruit was fully ripe and most of it had fallen from the trees. During the intervening period twelve samples of fruit were taken, and the hormone content of the seed was determined for all but the first of these, when the seeds were too small to be handled. The results, together with data on fruit and seed weight, are presented in Table I and are shown in graphical form in Fig. 1.

TABLE I.

Data on growth of fruit and seed, and on the hormone content of the seed in the apple variety Beauty of Bath.

Date 1947.	Days after petal fall.	Fresh weight fruit (gm.).*	Dry weight seed (mgm.)†	μg. equivs. BNOA.	
				per 100 seeds.	per gm. dry wt.
May 29	12	0·54	0·18	0·1	5·6
June 3	17	1·2	0·44	0·3	6·8
,, 10	24	3·4	1·7	1·0	5·9
,, 17	31	7·6	3·5	15·2	43·5
,, 24	38	10·7	4·7	5·2	11·0
July 1	45	18·5	6·7	3·6	5·4
,, 8	52	21·6	7·4	7·7	10·4
,, 18	62	37·2	10·9	13·6	12·5
Aug. 1	76	59·2	19·7	41·3	21·0
,, 8	83	57·6	22·2	23·5	10·6
,, 15	90	60·4	24·9	6·8	2·7

* Mean of 25 fruits.
† Mean of 100 seeds.

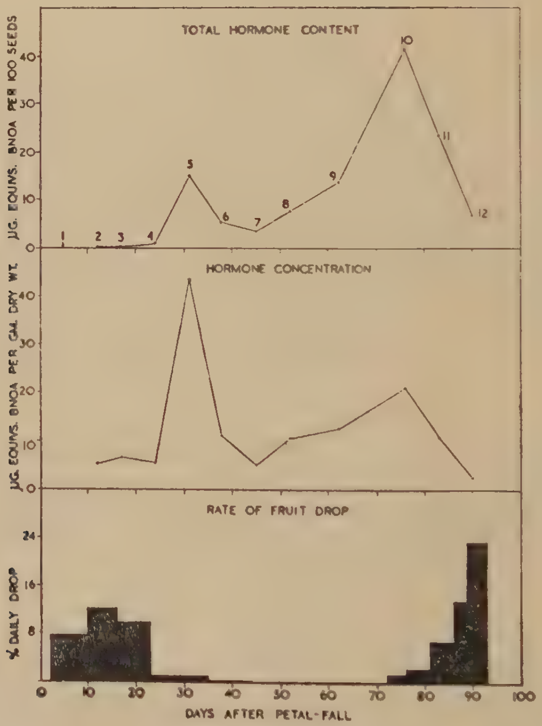


FIG. 1.

Hormone content of seed, concentration of hormone in seed, and rate of fruit drop for the apple variety Beauty of Bath. Season 1947.

Extracts prepared from seeds 12, 17 and 24 days after petal fall showed almost negligible activity when applied to tomato ovaries, even in concentrations as high as 1,000 seeds per ml. of extract. One week later, however (Fig. 1, Stage 5) a highly active extract was obtained. The hormone content of subsequent samples again fell until the 45th day (Fig. 1, Stage 7), after which it rose steadily to a maximum on the 76th day. This second peak on the curve corresponded closely to the date at which this variety would normally have been picked for market. Fruit on the experimental trees, however, was not picked and two further samples were taken at eighty-three and ninety days after petal fall. These showed that, in the final stages of maturity, there was a very rapid drop in the hormone content of the seed.

The curve showing the concentration of hormone in the seed, expressed on a dry weight basis, also shows two maxima but owing to the great increase in seed weight between the 31st and the 76th days, the second peak is not so well marked as in the curve showing total hormone content.

SEED AND FRUIT DEVELOPMENT.

Successive stages in the development of the fruit and the seed are shown in Fig. 2. The general course of development is similar to that described for other apple varieties by previous workers (7, 9), and three main phases can be recognized, viz. :

Phase 1. (Fig. 2, Stages 1-4.) Characterized by a rapid increase in the size of the seed and of the nucellus. The embryo develops only very slowly, but the triple fusion nucleus undergoes many divisions, furnishing the embryo sac with a large number of free endosperm nuclei. The fruit shows a rapid swelling. In Beauty of Bath this phase was found to last until about twenty-five days after petal fall.

Phase 2. (Fig. 2, Stages 5-10.) The endosperm becomes cellular and shortly afterwards the embryo enters on a period of rapid growth. During this phase the endosperm is continually being broken down and absorbed by the growing embryo on the inside, but is continually regenerated by meristematic activity around its periphery, and actually increases considerably in volume. The tissues of the nucellus are also broken down but, unlike the endosperm, are not regenerated, so that by the end of this phase the nucellus has almost disappeared. The fruit continues to grow and approaches its maximum size. The duration of this phase was found to be about fifty days.

Phase 3. (Fig. 2, Stage 12.) The embryo is now fully grown and the seed turns from white to brown as the testa hardens. The endosperm ceases its meristematic activity and begins to degenerate. During this phase the fruit increases slightly in volume, ripens, and falls from the tree. In Beauty of Bath this was found to be the shortest of the three phases, lasting about fourteen days, by which time nearly all the fruits had dropped.

HORMONE CONTENT AND ENDOSPERM DEVELOPMENT.

Comparison of Figs. 1 and 2 shows that the sudden appearance of large quantities of hormone in the seed one month after petal fall (Stage 5) corresponded closely with the time of formation of the endosperm, details of which are shown in Fig. 3. Fig. 3a shows the embryo sac twenty-four days after petal fall (Stage 4), when the hormone content of the seed is still very low. At the micropylar end can be seen the young

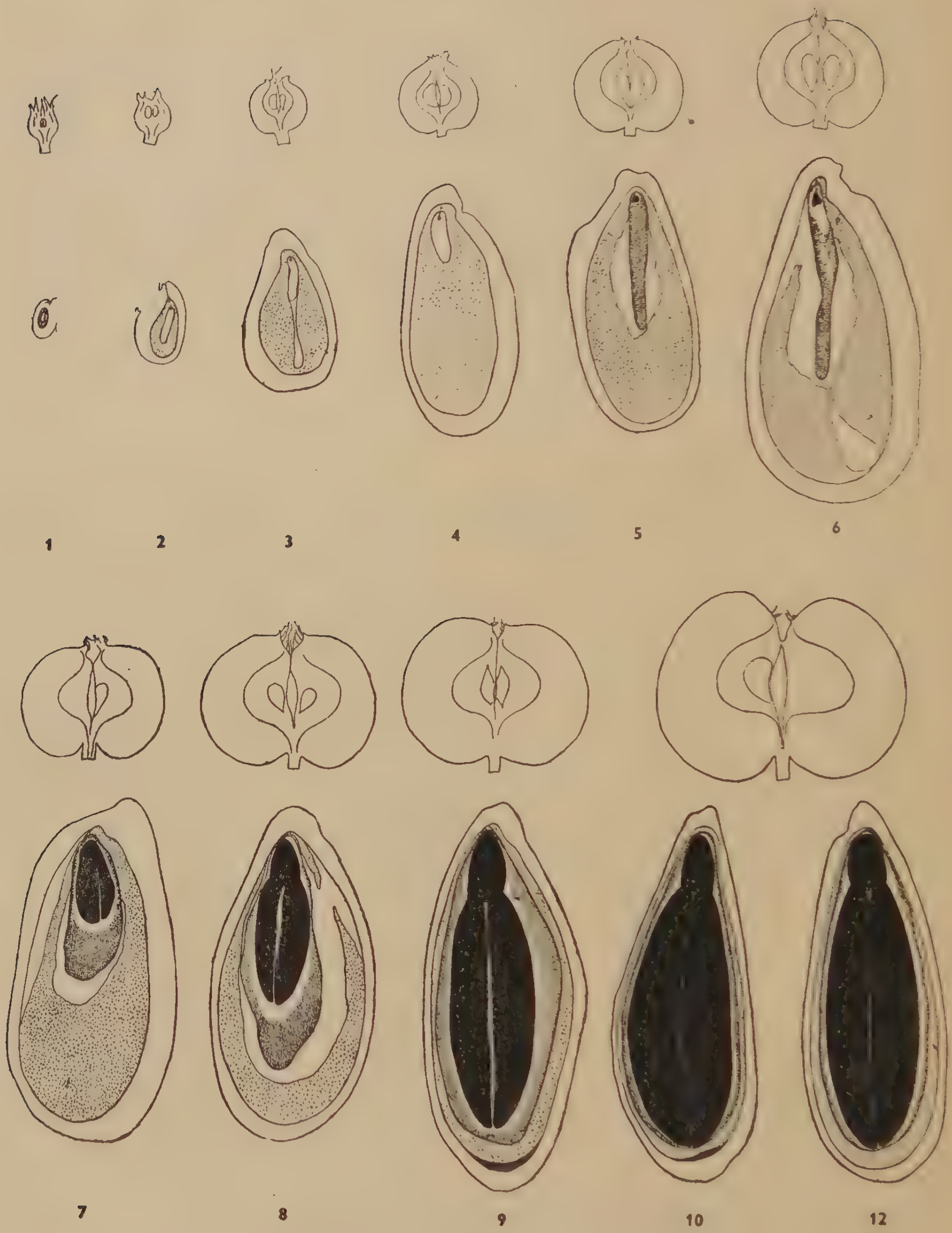
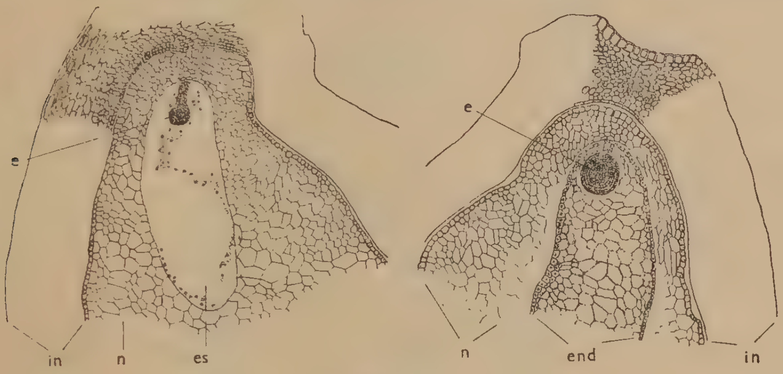


FIG. 2.

Fruit and seed development in the apple variety Beauty of Bath. The numbers of the developmental stages correspond with those on the upper graph in Fig. 1. The lightly stippled tissue in the seed is nucellus, the heavily stippled is endosperm, and the black, embryo. Magnification : Fruits × ½. Seeds × 7.

embryo and scattered throughout the embryo sac are numerous free endosperm nuclei connected by strands of protoplasm. Fig. 3b shows a similar embryo sac one week later (Stage 5), when the hormone concentration in the seed is at the maximum. The embryo is still quite small and the cotyledons have not yet differentiated, but it will be observed that the embryo-sac is now filled with cellular endosperm tissue and that the inner layers of the nucellus have already started to disintegrate. The endosperm cells of the peripheral layer are smaller than the rest and have large nuclei and dense, deeply staining cytoplasm; they constitute a meristematic zone by which the endosperm grows to accommodate the rapidly enlarging embryo.

The close correlation between the time of formation of a cellular endosperm and the appearance of marked activity in the seed extracts, strongly suggests that the endosperm itself might be the seat of production of this hormone. Confirmatory evidence of this was obtained for seeds of two other apple varieties collected at later stages of development when the embryo was almost fully grown. In these older seeds it was found possible to separate the embryo from its enveloping layers of endosperm and nucellus and to determine their hormone content separately. The results, given in Table II, show that in Miller's Seedling the concentration of hormone in the outer layers (endosperm and nucellus) was approximately eighteen times as great as in the embryo. Since, in these seeds, the nucellus had been almost completely absorbed, there seems little doubt that the bulk of the hormone was present in the endosperm tissue. The Crawley Beauty seeds were sampled at an earlier stage of development before the nucellus had completely disintegrated but, here again, the bulk of the hormone content of the seed was found to reside in the outer layers. These results support the view that the hormone originates in the endosperm and that the small amounts found in the embryo arrive there by diffusion.



a. FIG. 3. *b.*

Details of endosperm formation. Left: twenty-four days after petal fall. Right: thirty-one days after petal fall. e = embryo; end = endosperm; n = nucellus; es = embryo sac; in = integuments. Magnification: × 32.

If this hypothesis as to the origin of the hormone is accepted it is pertinent to enquire why, once the endosperm has become organized as a cellular tissue, the hormone content of the seed again falls to a low level (Fig. 1, Stages 6 and 7), particularly as the endosperm itself shows a marked increase in volume (Fig. 2).

A possible explanation of this is that the hormone plays some part in embryo development and, during the period of rapid growth, is used up more quickly than it is produced by the endosperm. That this is actually so seems to be indicated by the fact that once the embryo has reached its full size, the hormone content of the seed again rises to a high level (Fig. 1, Stage 10). The condition of the endosperm at this stage of development is shown in detail in Fig. 4a. Although largely obliterated by the growth of the embryo there are still four to six layers of active endosperm cells remaining. These have large nuclei and dense, deeply staining cytoplasm; their arrangement in radial rows suggests recent meristematic activity. The total volume of active endosperm tissue in the seed at this stage of development was estimated at about 2 cu. mm. The initial volume of the endosperm at Stage 5 was estimated to be about 0·6 cu. mm., so that the total hormone content of the seed at Stages 5 and 10 (Fig. 1) was roughly proportional to the volume of active endosperm tissue present.

TABLE II.

Hormone content in different parts of the seed.

Variety and date.	Part of seed.	Av. dry weight (mgm.).	μg. equivs. BNOA.	
			per 100 seeds.	per gm. dry wt.
Miller's Seedling 7/8/47	Embryo	10·0	15·2	15·2
	Endosperm and nucellus	2·0	55·4	277·0
Crawley Beauty 12/8/47	Embryo	8·7	0·8	0·9
	Endosperm and nucellus	3·4	2·4	7·1

During Phase 3 the remaining endosperm tissue showed a rapid deterioration and Fig. 4b shows the condition of this tissue in the mature seed (Stage 12). The cells are no longer arranged in distinct radial rows and the protoplasmic contents have shrunk away from the cell walls. This final deterioration of the endosperm appeared to be correlated with the rapid drop in the hormone content of the seed between Stages 10 and 12 (Fig. 1).

RATE OF FRUIT DROP.

The average percentage daily drop of fruits for successive periods was calculated by expressing the total fruits dropped during the period as a percentage of the number of fruits on the tree at the beginning of the period, and then dividing by the number of days. The results are shown in graphical form in the lower part of Fig. 1.

The post-blossom drop was exceptionally heavy and for the first twenty-three days after petal fall the young fruitlets dropped at the average rate of 8 per cent. per day. The cause of this heavy drop was not inadequate pollination, for the large majority of the dropped fruitlets were found to contain well developed seeds, and were growing rapidly at the time of abscission. The fruitlets dropped between the 16th and 23rd day, for instance, averaged $\frac{3}{4}$ to 1 inch in diameter.

By the 23rd day after petal fall, the fruitlets remaining on the tree represented only 3·5 per cent. of the number of blossoms. At this point the drop practically ceased and the rate of dropping remained at a very low level until about eighty days after petal-fall, when the pre-harvest drop started. The usual intermediate or "June" drop was not observed in this variety in 1947.

The striking correspondence between the appearance of the hormone in the seed and the cessation of the post-blossom drop (Fig. 1, Stage 5) strongly suggests that the hormone itself is responsible for inhibiting the development of the abscission layer at the base of the fruit stalk. The final rapid fall in the hormone content of the seed also seems to correspond well with the occurrence of the pre-harvest drop, though the absence of an intermediate drop at Stage 7, when the hormone content of the seed reached a fairly low level, is difficult to explain.

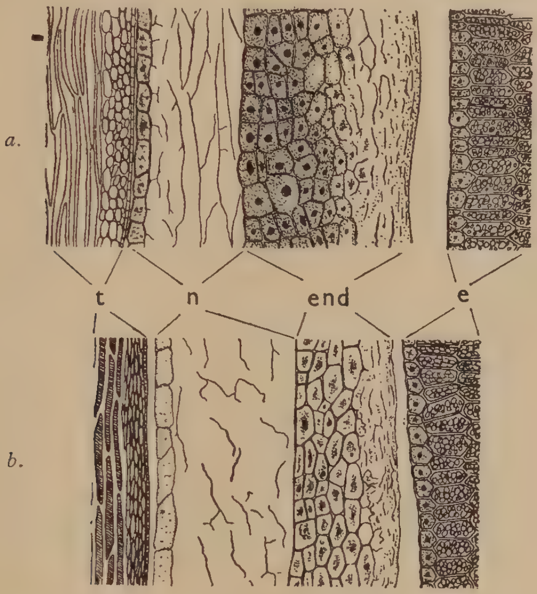


Fig. 4.

Longitudinal sections through the outer parts of the seed showing the condition of the endosperm at seventy-six days (above) and ninety days after petal fall (below). end = endosperm; n = nucellus; e = embryo; t = testa. Magnification: × 120.

Further evidence that the hormone content of the seed may be a factor in controlling fruit drop was obtained for two other varieties of apple which showed a typical "June drop", viz. Cox's Orange Pippin and Miller's Seedling. A comparison of picked and dropped fruits collected on the same day showed that not only did the dropped fruits contain fewer seeds on the average than the picked fruits, but also the hormone content of the seeds was considerably lower (Table III).

Effect on petiole abscission.

If the function of this hormone, as seems to be indicated, is to control fruit abscission, it should also be active in delaying the abscission of de-bladed petioles of such plants as Coleus and Ivy, in which the mechanism of abscission is similar to

TABLE III.

Fruit weight, seed number and hormone content of seed of picked fruits and "June drop" fruits in the apple.

Source of fruit.	Av. fruit weight (gm.).	Av. no. seeds per fruit.	µg. equivs. BNOA per 100 seeds.	µg. equivs. BNOA per fruit.
Cox's Orange Pippin. July 15th.				
Picked	21·2	6·0	12·3	0·74
Dropped	11·2	4·4	5·6	0·25
Miller's Seedling. July 16th.				
Picked	24·3	7·4	18·2	1·35
Dropped	15·1	6·5	4·9	0·32

that involved in the pre-harvest drop of apples. In order to test whether this was so, the following experiment was carried out :

50 gm. of dried and powdered apple seeds of the varieties Miller's Seedling and Crawley Beauty, collected at various stages of development, were refluxed for one hour with 250 ml. of distilled water. After separation from the plant material the water extract was washed three times with 400 ml. peroxide-free ether and the ether washings taken to dryness in a small conical flask. The residue was dissolved in 7 ml. hot water and allowed to cool. A dense precipitate of phloridzin was filtered off and the filtrate washed three times with 50 ml. ether. The ether extract was taken to dryness in a small specimen tube and the residue (0·038 gm.) dissolved in 0·2 ml. water and mixed into an emulsion with 0·56 gm. lanolin.

One-year-old shoots of Ivy (*Hedera helix*) were collected and stood with their bases dipping into tubes of water. The petioles of 100 leaves were severed at a point ¼ inch from the stem and the cut ends were smeared with the lanolin emulsion containing the seed extract. Another series of 100 petioles were treated with pure lanolin.

The results of this experiment, presented in Table IV, confirm that the seed extract was highly active in delaying abscission. The same extract applied unilaterally to young internodes of the tomato stem, however, failed to produce any curvatures, so that there is no evidence that this hormone is in any way active in stimulating cell extension.

TABLE IV.

Effect of apple seed extract in delaying the abscission of Ivy petioles. Petioles treated on January 12th, 1948.

Treatment.	No. petioles treated.	No. of petioles abscissed after :—						
		10 days.	15 days.	20 days.	25 days.	30 days.	35 days.	40 days.
Lanolin only	100	3	34	80	96	100	100	100
Apple seed extract	100	0	0	0	7	45	92	100

Phloridzin content of seed.

The crystalline precipitate which is formed when a hot water extract of apple seeds is cooled has been identified as phloridzin (10), a glucoside which was previously known to occur in the root- and stem-bark of the apple (3). The weights of the crude phloridzin residues extracted from seeds at different stages of development, and the concentration of phloridzin in the seed, are shown in Fig. 5. The curves show a general resemblance to those for hormone content and concentration, with the exception that the highest concentrations of phloridzin (14·5 per cent. of the dry weight) occur in the very young seeds, before the hormone has appeared and before the endosperm tissue has been formed. In experiments in which the embryo and the outer layers of the seed were extracted separately, phloridzin was recovered only from the outer layers. These observations suggest that phloridzin is present in the nucellus and probably in the endosperm as well, and that its role is that of a reserve food material which can be rapidly hydrolyzed to supply the growing embryo with glucose.

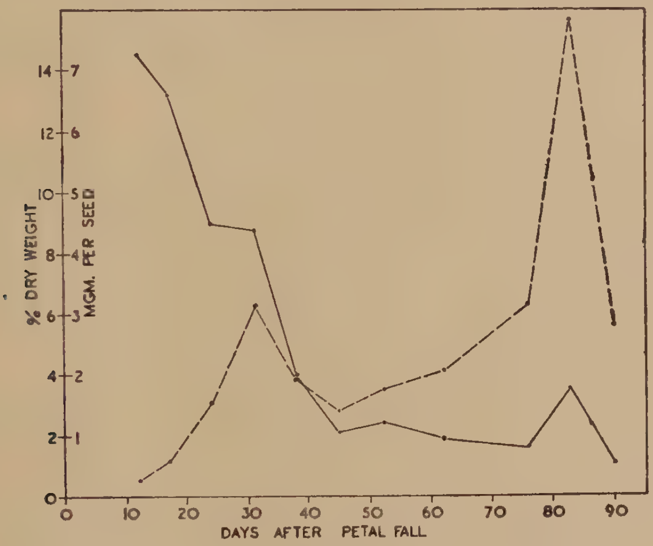


Fig. 5.

Phloridzin in the seed as (*a*) percentage of dry weight (solid line) and (*b*) mgm. per seed (broken line).

The possibility that one of the breakdown products of phloridzin might actually be the active compound was investigated, but both phloretin and phloretic acid were found to be without activity when tested on tomato ovaries.

DISCUSSION.

Relation between hormone and growth of fruit.

There is a considerable body of evidence supporting the view that the growth of apple, pear and other fruits is under the control of a hormone produced in the seeds. Kobel (4), for instance, has demonstrated a significant correlation between seed number and fruit weight in the apple, and Brittain and Eidt (1) have shown that lack of symmetry in the fruit is due to low seed development. Similar relationships for the pear have been reported by Rodrigues (8). Kobel has also shown that the sugar and acid content of the fruit may be influenced by the number of seeds.

On the other hand, the hormone which is extracted from apple seeds by boiling water, although active in inducing parthenocarpy in the tomato, appears to play no direct part in stimulating fruit growth in the apple. There are two observations which lead to this conclusion : (1) For the first twenty-five days after petal fall there is practically no hormone present in the seeds, yet during this time the young fruitlets swell rapidly and attain a diameter of approximately 1 inch. (2) There is no correlation between the growth rate of the fruit and the hormone content of the seed. The growth of the fruit can be represented by a smooth sigmoid curve (Fig. 6) which in no way reflects the wide fluctuations in hormone content at different stages of growth.

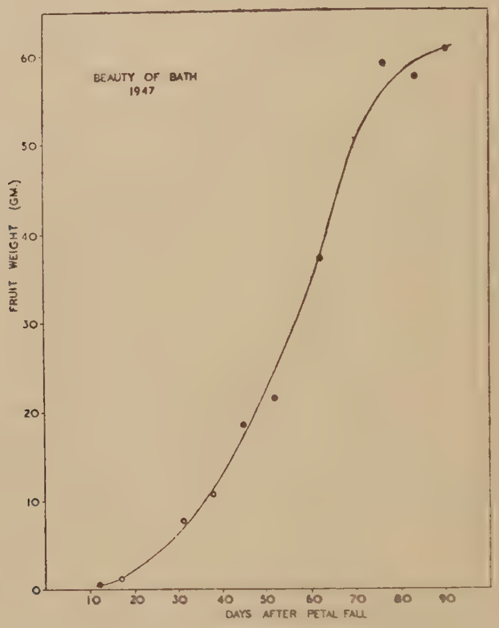


Fig. 6.
Growth curve of the apple variety Beauty of Bath throughout the season 1947.

There are several possible explanations of these inconsistencies. Fruit growth may, for example, be under the control of a second seed hormone which either is not extracted by the hot water method or is not active in the tomato ovary test used in this investigation. Alternatively the fruit-growth hormone may not have its origin in the seed, but may enter the fruit through the pedicel, or be formed in the stylar tissue under the stimulus of pollen tube growth, or even come directly from the pollen tubes themselves. Gustafson (2) and Yasuda (11) have demonstrated that in some plants, the pollen contains hormones capable of stimulating parthenocarpic development of the fruit, though in the apple it is doubtful whether pollen tube growth alone, unaccompanied by fertilization, is ever effective in initiating fruit development.

Although the hormone which has been extracted from the endosperm does not appear to play any direct part in stimulating fruit development, it does seem to be indirectly essential in so far as it prevents abscission of the developing fruit. It may, in addition, play some vital role in embryo growth.

The close relationship between endosperm development and fruit abscission, here shown to occur in one variety of apple (Beauty of Bath), seems to be a feature which has been overlooked by previous investigators, but should further work confirm this relationship for other varieties, it might provide a new approach to the problem of fruit drop and its control.

SUMMARY.

1. Previous work had shown that it was possible to extract from apple seeds at certain stages of development a hormone which was active in stimulating parthenocarpy in the tomato.

2. Using a hot-water extraction technique and a quantitative form of the tomato ovary test, measurements were made of the quantity and the concentration of this hormone in the seed of the apple variety Beauty of Bath at approximately weekly intervals from petal-fall to fruit ripening.

3. The hormone first appeared in the seed in large quantities thirty days after petal-fall, its appearance coinciding with the formation of the endosperm and with the cessation of the post-blossom drop.

4. Following endosperm formation there was a period of rapid embryo growth, during which the hormone content showed a temporary reduction, rising again to its maximum level seventy-five days after petal-fall, when the embryo had attained its full size.

5. In the final stages of fruit growth there was a rapid fall in the hormone content of the seed, which appeared to be correlated with the degeneration of the endosperm and the occurrence of the pre-harvest drop.

6. In Cox's Orange Pippin the hormone content of seeds from " June drop " fruits was 45 per cent. of that from picked fruits. In Miller's Seedling it was only 27 per cent. In addition, the dropped fruits had fewer seeds on the average than had the picked fruits.

7. The highest concentrations of hormone were found in the outer layers of the seed and it is concluded from this and other evidence that the hormone originates in the endosperm.

8. This hormone appears to play no direct part in stimulating the growth of the fruit, and its chief function is thought to be the control of fruit abscission. It is shown to be active in delaying the abscission of shortened petioles of Ivy.

9. The glucoside phloridzin was found in large quantities in the outer layers of the seed and is probably present both in the nucellus and in the endosperm. Its function appears to be that of a reserve food material.

ACKNOWLEDGMENTS.

I wish to thank the many helpers who, from time to time, assisted with the tedious task of extracting seeds from the apples, and especially Miss B. Febrey and Dr. D. Woodcock, who was also responsible for the preparation of phloretin and phloretic acid.

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(*Received* 26/2/48.)

STUDIES IN BIENNIAL BEARING

II. A REVIEW OF THE LITERATURE

By L. B. SINGH
East Malling Research Station

In the first paper of this series (Hoblyn, Grubb, Painter and Wates, 1936), it was stated that a Bibliography on biennial bearing would be prepared. Owing to various circumstances, this has not proved possible until the present moment and this review is now submitted as an introduction to a new series of studies on the subject.

Biennial bearing, i.e. alternate year cropping, of certain fruit plants is a problem of great economic importance in commercial fruit growing all over the world. Trees which have acquired this biennial rhythm carry a heavy crop in one year (called the " on " year) and little or no crop in the next (the " off " year). The fruits in the " on " year tend to be overcrowded, small and of poor quality, while the few fruits that are formed in an " off " year are usually above normal size and often too large for the best commercial purposes. The alternation of too much and too little crop, in the " on " and " off " years respectively, may persist with great regularity, though it may be upset by some major climatic factor (such as a severe spring frost in the " on " year).

Biennial bearing has been reported from many countries and in many kinds of fruit. In England it is especially serious in the apple, though it is also serious in pears and occurs in certain other tree fruits. It is a markedly varietal characteristic, some varieties suffering much more seriously than others, as will be discussed later. Many workers from the U.S.A. and Canada have reported the biennial habit in different varieties of apples and pears. Reports from Australia include those of Carne (1931) and Bowman (1932) discussed below. Hamilton (1932) from New Zealand, Blinov (1938) from the U.S.S.R. and workers from other apple growing regions of the world have also reported the biennial habit of apple varieties.

In citrus fruits the biennial habit of Valencia orange has been reported by West, Barnard and Allan (1937) in Australia, and by Hodgson and Eggers (1941) in America. Brichet (1937) reported the biennial habit of certain citrus varieties in Morocco.

Among other crops, it is well known that many varieties of mango, one of the chief fruits of tropical and sub-tropical regions, grown in India, Java and Egypt, bear a profitable crop only in alternate years. Recently, the importance of the problem has also been realized with regard to coffee, Gilbert (1935) having reported biennial bearing in coffee in East Africa. Beaumont (1939) and Pais (1943) observed similar behaviour of coffee trees in Hawaii and India respectively. Anagnostopoulos (1937) and De Almeida (1940) reported alternate year cropping of olive trees in Greece and Portugal respectively.

THE LITERATURE

The literature covering biennial bearing is extensive and no comprehensive review of it has hitherto appeared. In this review the literature will be considered under the following headings :—(*a*) On economic importance ; (*b*) on measurement of biennial bearing ; (*c*) on causes of biennial bearing ; (*d*) on attempted control.

(*a*) ON ECONOMIC IMPORTANCE.

The economic effect of this trouble is by no means limited to variation in the yearly crop on individual fruit farms. Large areas are often in the " on " or " off " year at the same time.

The price realized by the sale of the produce in the " on " years is low as the markets tend to be glutted by the abundant crop. while in the " off " years the growers have little crop to offer although the prices are high. The marked annual variations in yield obviously cause an uneconomic use of plant and equipment, e.g. spraying, packing and storage machinery, and lead to high overhead expenses.

The marked fluctuation in crop yields from year to year affects not only the growers but all those who are engaged in the industry, e.g. agents and distributors, shipping companies and manufacturers, whose business is affected by the lack of continuity of supply.

Specific work on the economic aspect of the problem includes that of Carne (1931), who made a short survey of the influence of heavy and light cropping in alternate years on the acreage of apple orchards in Australia. His survey showed a decline in planted area which he considered was mostly due to the marked fluctuation of production in alternate years.

Bowman (1932) pointed out the disastrous effects of alternate cropping on the apple industry in New South Wales. He observed that biennial bearing seriously upset the price stability and market organization and quoted the report of the first Bathurst (N.S.W.) conference of producers and consumers which stated that apple growers of all the primary producers received the lowest net profit for their produce. Apple growers received only 27 per cent. of the retail price ; pear and orange growers 34 per cent., while maize growers got 55 per cent. and wheat growers 83 per cent. This he attributed to the irregular and unreliable supply of the fruit crops. Bowman (1940) found that the yield from two consecutive " regular " crops was 50 per cent. higher than that from one heavy crop followed by an "off" year crop. Wilcox (1944) also showed that the biennial habit resulted in a considerable reduction in crop per tree in the Okanagan Valley of British Columbia.

Thus it is no exaggeration to say that biennial bearing is one of the major problems of the fruitgrowing industry to-day.

(*b*) ON MEASUREMENT OF BIENNIAL BEARING.

Although the major features of the biennial habit in any given tree or set of trees can be readily seen by examining the crop figures for several successive years, this provides no numerical standard for making a detailed comparison between different sets of trees.

Such a basis of comparison is obviously most desirable, but it was not until 1936 that a method of numerical expression of biennial bearing was devised by Hoblyn, Grubb, Painter and Wates (1936). The first expression introduced by these authors, the factor " B ", indicates the extent to which the cropping performance of a tree is regularly annual, regularly biennial, or irregular. To determine " B ", crop records for at least three years must be available. The yield for each year is considered in relation to that of the previous year. If the second yield exceeds the first, the difference is given a plus sign and if the converse, a minus sign. Thus a series of signs for each consecutive pair of years is obtained. From these, " B " is calculated by determining the percentage of consecutive pairs of unlike signs over the whole period. A value of " B "=100 is obtained when the tree is completely biennial, i.e. when + is followed by –, and vice versa, throughout the period. On the other hand, when " B "=0 the tree is regularly increasing (or regularly decreasing) in yield.

The second factor, " I ", was employed as a measure of the intensity of degree of crop fluctuation from year to year. The derivation of " I " may be expressed in the form :

$$I=\frac{\text{difference between successive yields}}{\text{sum of successive yields}}$$

The values of " I " vary from 0 to 1, where 0 denotes equal crops in successive years and 1 no crop at all in alternate years. Values of " I " may be averaged for a given number of years and a mean value obtained.

Some possible modifications of the method of calculating " I " are described in the original paper.

By making use of the above two values, Hoblyn *et al.* compared some fifteen varieties of apple by their crop records at East Malling with regard to biennial tendency and found a number of them showing a strong biennial behaviour, e.g. Newton Wonder, Gladstone, Early Victoria and Allington. Bramley's Seedling, Miller's Seedling and Melba which are also well known for their biennial bearing habit were not included in the above survey. Some others, e.g. Cox's Orange Pippin and Norfolk Beauty, indicated irregular rather than biennial bearing. James Grieve and Worcester Pearmain gave lowest values both for " B " and " I " and therefore were classed as regular croppers. However, these authors concluded that varieties may behave very differently under widely different conditions. Other conclusions from this important study will be discussed later.

In mature trees of a variety known to have the biennial habit to a greater or less extent, the factor " I " can be used alone as a sensitive index of the effect of any given treatment. In fact Wilcox (1944) later used the " I " formula of Hoblyn *et al.* multiplied by 100 and called this figure the Biennial Bearing Index. Thus his

$$\text{" Biennial Bearing Index "}=100\ \frac{\text{(difference between two yields).}}{\text{(sum of two yields)}}$$

Complete annual bearing gave an index of zero and complete biennial bearing and index of 100. He also calculated in a similar way the Biennial Indices for increases in trunk circumference, terminal shoot length and twig weight and studied the correlations between these and the Biennial Bearing Index, thus using for the first time a new and convenient method of interpreting biennial bearing in relation to variations of tree vigour.

The Biennial Index was also used by Dickson (1946) in some cultural studies. There is no doubt that the introduction of these formulae is a very useful step forward in the study of the biennial habit.

(*c*) ON CAUSES OF BIENNIAL BEARING.

(1) *Fruit bud formation.* A useful chronological review of work on fruitfulness up to 1918 was made by Wiggans (1918). His paper showed that earlier workers devoted themselves mainly to the study of factors responsible for fruit bud formation, based on the assumption that a good show of blossom is the first essential for a good

crop of fruit. At the beginning of the investigations, some workers argued that the amount of blossom formed was inherited while others maintained the view that flowers were dependent upon the presence of a certain specific blossom building substance. Most of the workers were in agreement on the one point that formation of fruit buds was dependent upon a supply of reserve food material. Gourley (1915) reported that the spur possessing a fruit bud had a greater supply of starch than one without a fruit bud. He also found that the leaf area of a non-bearing spur was greater than that of a bearing spur, and considered that this larger leaf area aided the formation of fruit buds for the following year by supplying a larger reserve of food material.

(1)*a*. *Time of fruit bud formation.* Goff (1899-1901) was the first person to make a close study of the exact time of fruit bud differentiation and its development prior to opening. He concluded that there were two periods for fruit bud formation; the first in early summer and the second in early autumn. He also reported that buds began to differentiate as flower buds about the time when wood growth ceased. This he explained on the ground that the energies of the trees were used up by vegetative growth until cessation of shoot growth and only then were they available for the formation of fruit buds and maturing the wood and fruit. Morgan (1902) observed that blossom buds of apple did not develop to any appreciable extent from November 15th to March 1st; while Drinkard (1909-10) found that fruit buds of Oldenburg apple began to differentiate in the last week of June and did so for a long period, although they were mostly formed in early July.

Gibbs and Swarbrick (1930), studying fruit bud formation in apple, concluded that there was considerable variation with regard to the actual time of flower bud initiation depending upon climatic conditions, age of tree, rootstock, variety, cultural conditions and pruning treatment. Tufts and Morrow (1925) gave June 11th as the earliest date on which they were able to observe fruit bud formation. Barnard and Read (1932) reported that apple buds differentiated from five to six weeks after the spur leaves had reached full growth. The majority of workers are thus of the opinion that flower bud initiation may be expected to begin by the middle of June or early July and that conditions at that time are very important for any modification of blossoming performance and consequently fruiting habit of the tree.

(1)*b*. *Fruit bud formation in relation to shoot growth.* The relation of shoot growth to fruit bud formation has been widely investigated and, as mentioned above, it has been observed that the time of shoot and spur growth cessation is to some extent related to fruit bud formation. Yeager (1916), Roberts (1920), Hooker and Bradford (1921), Auchter and Schrader (1923) and McCartney (1925) have all shown that trees of regular bearing habit, whether of annual or biennial varieties, present a marked and positive correlation between length and diameter of spurs and blossom bud formation. In biennial bearing trees, this correlation could not be established, the heavy crop preventing the blossom bud formation, irrespective of the length of shoot growth. Swarbrick (1928) found that the earlier the shoots stopped their linear growth in the summer, the higher was the percentage of blossoming the following year. Warne and Wallace (1935) found a high negative correlation between the rate of shoot growth during certain periods in July and the percentage of fruit buds formed. This also fits in with the theory that growth requirements take priority over storage in the tree.

Barnard and Read (1932) found that fruit bud formation in pears coincided with cessation of growth of the current season's shoots ; Finch (1935), working with a number of biennial bearing apples, observed that terminal growth was first initiated and proceeded more rapidly in the fruitful than in the unfruitful shoots.

Barnard (1938) further stated that, in pome fruits, the time of bud differentiation seemed to be more closely related to the size of the current season's crop and to the time of cessation of shoot elongation growth than to climatic conditions. Micklem (1938) studied the time of fruit bud differentiation for apples as well as for certain other fruits such as pears, plums, apricots and peaches and concluded that a rigid date for the change of a vegetative bud into a fruit bud could not be given. However, he also found that under local conditions fruit bud differentiation started shortly after the cessation of growth in length of shoots.

Harvey (1923), Proebsting (1925) and Roberts (1926) all considered that the linear growth of terminal shoots, which was closely related to the formation of fruit buds, was mostly made at the cost of stored up food and was therefore naturally affected by the growth and fruiting conditions of the previous year. Mack (1924), however, reported that there was longer terminal growth, as a rule, in the " on " year than in the " off " year. Tucker and Potter (1929) confirmed this and Roberts (1926) suggested that shoots did not grow enough in length and diameter in the " on " year to form blossom buds in that year. Thus, a quantitative relationship between shoot growth and blossom bud formation does not seem to be clear.

(2) *Tree vigour*. One of the most commonly accepted theories is that the tree needs rest after carrying a heavy crop. In the " on " year it becomes exhausted owing to the big demand on its reserve by an overwhelming number of developing fruits and is thus incapable of storing enough reserve for laying down a sufficient number of fruit buds for the following year. The drain on its resources is so heavy that the tree in the subsequent season is incapable of setting even the few blossoms that may be on the tree, consequently the tree bears little or no crop. In this year fruit bud formation is, however, greatly favoured and the following year again a heavy crop of fruit is borne. Thus the biennial rhythm sets in and continues.

Pickering (1905, 1907, 1916) noted that fruiting may greatly reduce the growth of trees. Auchter (1917) emphasized the fact that heavy cropping may be injurious to trees, especially young ones, by causing weak growth generally. Partridge (1919) reported that trees which were cropping in alternate years increased in trunk circumference more in the " off " year than in the " on " year. Roberts (1926) and Finch (1927) corroborated the above finding by anatomical studies. Chandler (1925) also reported that at Cornell University, measurements of a number of trees showed that the annual increase in trunk tends to be strikingly reduced in the year when the tree is bearing a heavy crop. Thus trees cropping biennially might be expected to show a negative relationship between amount of crop and increase in trunk circumference.

Wilcox (1937) made an extensive study of the relationships between growth and fruiting of apple trees by the use of scatter diagrams and correlation coefficients and produced interesting new data among which were some relating to growth performance of biennial bearing trees. He observed that increase in trunk circumference was greater in the " off " year but terminal length was more in the " on " year. In a further study (1944) in which he recorded a series of 290 McIntosh trees selected from commercial orchards in the Okanagan Valley of British Columbia, he correlated biennial bearing index with the biennial trunk-growth index, the biennial terminal-growth and the biennial twig-weight index (the latter three as indicators of tree vigour) and found all of these correlations to be highly significant. He thought that biennial bearing was more closely associated with meteorological conditions than with lack of tree vigour.

(3) *Climatic effects.* The effects of various climatic factors such as rain (as affecting both air humidity and soil moisture) and frost in fruit setting and biennial bearing have also been well recognized. Pickering (1916) reported that the weather of England tended to form a biennial cycle which he considered to be associated with biennial bearing of dwarf trees. Macoun (1917) offered the same suggestion regarding alternate bearing in Canada. However, the meteorological records in England do not confirm any such biennial cycle in climate. Hedrick (1908), Gourley (1915), Auchter and Schrader (1933) and Potter (1937) have all stated that frosts may be the factor which starts biennial bearing of apple. It has been demonstrated only too often in England in recent years that spring frosts, destroying the fruit blossom, can throw trees into an " off " year condition, in which an excessive amount of fruit bud is laid down, resulting in a heavy " on " year following and thus initiating a biennial rhythm.

Lees (1926) working at Long Ashton found that a wet summer was followed by low fruit bud formation and therefore by low fruit yield and a dry summer by a heavier crop. This he attributed to the influence of rainfall on the internal condition of the tree, i.e. on the quantity of food material stored in the tree during the vegetative period.

Collison and Harlan (1927) suggested that the period of about six weeks from July 16th to September 1st was the critical period for moisture in an orchard of Rome Beauty apple trees. They found that any departure from normal rainfall affected the annual crop yield. They found, however, no such correlation between temperature and crop yield.

Aldrich and Work (1934), Magness (1934) and Degman, Furr and Magness (1933) also reported increased blossom bud formation due to prolonged periods of early summer drought.

Auchter, Schrader, Laggasse and Aldrich (1926) and Paddock and Charles (1929) stated that shading restricted fruit bud formation as it reduced the supply of carbohydrates. This could not, however, normally be a major factor in bringing about biennial bearing. Thus it appears that climatic effects, though they may initiate or modify biennial bearing, are not a major factor in its continuance.

(4) *Spur behaviour.* Roberts (1920) attributed biennial bearing neither to climatic conditions nor to overbearing but to over-blossoming which in itself is an effect of nutrition, stating that " nutritional conditions which cause formation of an extreme number of blossom buds tend to give ' off ' year bearing ".

Poljakov (1941) also observed that alternation of high and low yield of fruit was due to wasteful expenditure of nutrients which, instead of being utilized by the tree for steady fruit bearing from year to year, go to form fruit buds in numbers far in excess of the number required for an adequate yield of fruit.

Some investigators have reported a close relationship of alternate bearing with the nature of growth and spur habit of biennial bearing trees. Butler (1917) remarked that, with few exceptions, spurs in apples bore flowers in alternate years and, in mature trees which form new growth slowly, a heavy crop will naturally be followed by a light crop. Roberts (1920) also reported that spurs which blossomed and set fruit remained vegetative in the following year and spurs which did not blossom, or even blossomed but did not set fruit, carried fruit the next year. Swarbrick (1933) at Long Ashton, also noted the marked biennial performance of individual spurs of Lane's Prince Albert apple.

This theory of individual spur behaviour does not, however, satisfactorily explain the biennial bearing tendency in young and vigorously growing apple trees nor, for instance, in mangoes which do not form spurs and yet are marked biennial bearers.

Chandler (1925) noted the biennial habit in certain varieties of apples at Cornell, at quite an early age, when they were still growing vigorously and producing new spurs each year. He concluded, therefore, that the biennial habit is not merely a matter of morphology, but is partly due to some physiological influence affecting the whole tree.

Further, apart from the individual biennial performance of spurs, Roberts (1920) reported a relation between spur growth and annual bearing. Hooker (1925), however, did not confirm this and concluded that the tree behaved as a unit determining the performance of spurs. Auchter (1917) also remarked that it was very seldom that spurs fruit one year and bloom the next, although mere blooming had only slight influence on the behaviour of the spur.

(5) *Physiological factors.* Lately, the problem of biennial bearing has been approached through physiological and chemical studies of tree tissues. The determination of nutritional conditions of bearing and non-bearing spurs as aiding or inhibiting fruit bud formation has formed the chief subject of study.

How a heavy blossoming, followed by a heavy crop of fruit, inhibits fruit bud formation has been explained physiologically by some investigators by the theory of carbohydrate-nitrogen ratio. Chandler (1925) suggested that the trouble may be due to exhaustion of one or more of the mineral nutrients; a reduction in the supply of carbohydrates generally, or of some particular carbohydrate or carbohydrate-nitrogen compound; depletion of some substance normally present in small quantities but necessary for fruit bud formation; development in the tissue of a general chemical condition unfavourable for fruit-bud formation or an unfavourable carbohydrate/nitrogen relationship. Kraybill and others (1925) advanced the theory that a proper balance of carbohydrates was the determining factor in bud formation. Summers (1924), Hooker (1930) and Murneek (1928) reached the same conclusion.

It is certainly a fact that, as shown by the older work of Gourley (1915) and the present author's work, discussed later, trees have plenty of leaves in their " off " year, which are likely to form a high quantity of carbohydrates, thus favouring fruit bud formation. In the " on " year the leaf area is relatively lower, as a result of which fewer carbohydrates are formed.

Roberts (1920) and Swarbrick (1928) showed that the leaf area on spurs was definitely related to bud formation and fruit setting. The significance, for fruit bud formation, of the presence of a greater leaf area than is needed for merely maintaining growth of fruits and trees, was also emphasized by Aldrich (1932), Magness, Overley and Luce (1932) and Harley, Masure and Magness (1933).

(6) *Chemical factors.* A chemical basis of alternate cropping was first investigated by Hooker (1922) who concluded that in apple trees, carbohydrate was deficient in the " on " year and nitrogen in the " off " year and also that these deficiencies robbed the spurs of their individual blossoming responses and caused them to

blossom or not to blossom as a whole. He also reported that the starch/nitrogen ratio was a better indicator of fruitfulness than the carbohydrate/nitrogen ratio.

The chemical investigations of Archbold (1928) showed that the greatest demand of the tree for nitrogen was during the blossoming and fruit setting period and of carbohydrate from then on until the fruit was practically full grown. However, Potter and Phillips (1930) also found no distinct relationship between carbohydrate nitrogen ratio and fruit bud formation. They found that blossom bud formation was constantly associated with the carbohydrate/insoluble-nitrogen ratio. They noted a significant difference in the content of relatively inert substances such as starch and recorded such differences as being at a maximum relatively late in the season.

Davis (1931) working with sugar prunes and Thomas and Barnard (1937) working with sultana, also reported that a high starch content appeared to be correlated with the formation of blossoms.

Harley, Magness, Masure, Fletcher and Degman (1942) made detailed investigations into the chemical composition of bearing and non-bearing spurs of apple trees : and reported that their separate morphological components differed widely in composition. They, too, observed a much higher starch percentage and a slightly lower one of free reducing substances in the tissues initiating blossom buds than in similar tissues forming leaf buds.

Bowman (1941), however, found with sugar prunes that the early stages of fruit growth, up to thirty-five days from the time of fruit setting, did not influence the composition of the tree but that it was the continued growth of the fruit that brought about changes in composition. Platenius (1932) also stated that formation of reproductive organs in celery was not determined by nutritional factors that involve the whole plant but probably by the localized accumulation of some chemical compounds in meristematic cells. He thus suggests that differences in chemical composition may not be the cause of differences in fruit bud formation, but are associated phenomena due to the same physiological causes.

Magness, Batjer and Baynes (1943) attempted to influence flower bud initiation in apples by using lanolin and other chemical growth substances, e.g. naphthalene-acetic acid, naphthalene acetamide in various strengths on various parts of spurs but failed to get any conclusive result.

(*d*) ON ATTEMPTED CONTROL MEASURES.

Repeated efforts have been made by many workers in the U.S.A. and England to find satisfactory control methods for biennial bearing. Most investigators have tried one or more of the following remedial measures :

1. Fruit thinning.
2. Blossom thinning.
3. Pruning.
4. Nitrogen manuring.
5. Defoliation.
6. Conservation of soil moisture at critical times.
7. Ringing.

Fruit thinning. This has been suggested as a means of reducing the exhaustion of the tree due to overcropping, so as to induce the formation of blossom buds in the fruiting year of biennial bearing. In some cases, thinning, practised every year, has afforded a considerable measure of success in preventing biennial bearing while in others no conclusive results were obtained. Gourley (1915), Auchter (1920), and Roberts (1920) all remarked that fruit thinning, though conducive to size improvement and quality of immediate crop, failed to bring about annual cropping in the trees of biennial bearing varieties.

Aldrich (1932) and Haller and Magness (1933) stated that thinning trees heavily so as to provide a comparatively large leaf area per fruit would tend to cause annual bearing. Harley, Masure and Magness (1934) also emphasized the importance of a high leaf/fruit ratio to annual bearing. In a further experiment (1935) they confirmed that biennial bearing of Yellow Newtown apple could be broken by heavy thinning, i.e. to a ratio of 70 leaves or 1,900-2,000 sq. cm. of leaf surface per apple. Tsuin (1941) however, concluded that the leaf/fruit ratio, which was chiefly concerned with carbohydrate supply, was not always the deciding factor for fruit bud differentiation, though he found that within a certain range there was an increase in fruit bud differentiation with increased leaf area, which differed from variety to variety. He also thought that varieties requiring a high level of leaf/fruit ratio were more strongly biennial.

Aldrich and Fletcher (1933) found fruit thinning rarely successful unless practised within six weeks of blossoming in apple. Dullum (1937) stated that fruit thinning could not influence biennial bearing unless it was practised too heavily, which meant the removal of at least nine-tenths of the fruitlets. Potter (1937) advocated early fruit thinning for overcoming the biennial bearing of McIntosh. Bowman (1940) remarked that fruit thinning could only be effective when only one fruit was left for every six spurs and if it was completed within five weeks after the date of full bloom. This he explained on the ground that there was a critical period extending from 30-35 days, during which alone a growing spur could lay down fruit buds, if not hampered by excessive fruit setting.

Fisher and Britton (1940), however, induced regular bearing in Newtown and Winesap apple varieties by extra heavy fruit thinning five weeks after full blossom. In the first year they thinned the fruit 20 in. apart and maintained it by thinning to one fruit per spur in subsequent years. Harley *et al.* (1942) established annual blossoming in field trials of biennial bearing varieties of apples by introducing earlier fruit thinning. They concluded that the severity of thinning necessary to influence blossom bud formation varied directly with tree vigour and the number of days after full bloom that thinning was performed. Fruit thinning, relatively heavy, performed early in the season, according to tree vigour, was likely to give the maximum fruit bud formation for the following season.

Fruit thinning in relation to blossom bud formation has also been closely studied with regard to certain other fruits. Tukey and Einset (1939) reported a marked increase in the number of blossoms in peaches in the season following fruit thinning.

Bowman (1941) reported similar results from his trials with sugar prunes and observed that at the critical time of blossom bud formation the fruit was in the period of rapid growth and suggested that this rapid rate of growth of an excessive crop at that time probably imposed an abrupt check on spur leaf growth which prevented a change in bud development that would culminate in blossom bud formation.

West, Barnard and Allan (1937) stated that early thinning, together with early harvesting, may induce regular bearing in Valencia orange. Hodgson and Eggers (1941) confirmed that early harvesting tended to give annual bearing in Valencia orange.

Recently much attention has been given to crop thinning or preventing fruit production altogether by application of sprays. Tar oils were tried at first but did not give satisfactory results. Howlett (1943) used di-nitro compounds for the same purpose and applied di-nitro-ortho-cyclo-hexyl-phenol and di-nitro-ortho-cresol to reduce fruit set, and found both of them quite effective in thinning the crop when used in oil emulsions. However, he advocated the use of sprays at full blossom rather than after fruit set, as much damage was done to the leaves.

Blossom thinning. While fruit thinning has been found to be only rarely successful in controlling biennial bearing, investigators have reported that blossom thinning is usually effective. Bailey (1929) stated that in the case of three apple varieties, namely Wealthy, McIntosh and Oldenburg, removal of between 75 and 100 per cent. of the blossoms may give a fair crop during the current season and yet allow the tree to lay down fruit buds for the following year. Read (1931) noted that typically biennial apples had been made to blossom heavily in the usual " off " year by removing their blossom in the preceding " on " year. Hoblyn *et al.* (1936) recorded the same effect of blossom removal at East Malling. Thies (1934) noted an increase in the leaf area by 41 to 54 per cent. by the end of May as a result of defloration in McIntosh apple, which he thought to be the cause of the increased fruit bud formation which appeared in the following year, and which normally occurred after defloration. Bobb and Blake (1939), however, attributed the success of blossom thinning in modifying the biennial bearing of Wealthy to several factors such as alteration of growth status, removal of terminal buds, etc.

The actual number of flower clusters that must be removed to make blossom thinning a successful operation, so that the tree may form fruit buds in the current season, has been found to vary from variety to variety, depending upon spur habit, tree vigour and other factors. McCormic (1933) recommended that two-thirds or three-quarters of the total spurs should be deflorated on Newtown whereas Bobb and Blake (1939) stated that blossoms should be thinned to 10 in. to 12 in. apart on Wealthy to enable it to blossom in the year following. Fisher and Britton (1940) concluded that blossom thinning which removed five out of six flower clusters in the pink bud stage, followed by fruit thinning to one fruit per spur five weeks later, would induce regular bearing in Winesap and Newtown. They remarked, however, that blossom thinning would fail if the tree was weak. They also noted that total yield was greatly increased on the trees converted from biennial bearing to regular bearing.

Blossom thinning, i.e. removal of superfluous fruit buds or flowers, has also been attempted by the use of sprays, as hand picking is a laborious as well as costly operation. Bowman (1940) used cresol at 1 per cent. emulsified with soap, tar oil at 2 per cent. and red oil at 4 per cent. for blossom thinning but reported only moderate success. Red oil was comparatively ineffective. Poljakov (1941) found Bordeaux mixture, 2 and 4 per cent. of green soap, iron sulphate and barium chloride as the most effective chemicals for the purpose. Childs and Brown (1942) reported modification of the biennial habit of Newtown by the use of tar oil at a concentration of 1 or 2 per cent., at a time when the trees were 10 per cent. in blossom. They noted a devastating effect on the young foliage but the trees recovered fully a few weeks later. They also recommended "spot spraying", i.e. drenching alternate branches rather than drenching the whole tree.

Recently, much work has been done on controlling fruit set, not only as a means of modifying biennial bearing but also of improving the current year's crop which, as a rule, in the "on" year of biennial bearing trees consists of very many small fruits. Auchter and Roberts (1935) tested a number of organic and inorganic spray materials for controlling the set of fruit. None of the inorganic materials, i.e. sodium polysulphide, copper sulphate, sodium nitrate, ferrous sulphate or sodium hyposulphite gave satisfactory results. Cresylic acid not only killed practically all the blossoms but also damaged twigs and spurs severely. They found tar oil distillates to be the most satisfactory spray, out of all the substances mentioned above, for the purpose, though it also did slight damage to twigs and leaves. Magness, Batjer and Harley (1940) reported promising results in changing the bearing year of biennial trees of York Imperial, Golden Delicious, Jonathan and Yellow Newtown by the use of tar oil distillates and DNO sprays. Magness and Batjer (1941) confirmed the effectiveness of tar oil distillate and di-nitro-ortho-cyclo-hexyl phenol when thoroughly applied during the cluster bud stage.

MacDaniels and Hildebrand (1940), for the first time, suggested Elgetol (sodium salt of di-nitro cresol) as a promising blossom spray for control of fruit set. Hoffman and MacDaniels (1941), and Hoffman (1942) also found Elgetol to be promising. Batjer and Moon (1943) succeeded in thinning Yellow Transparent by using Elgetol but remarked that factors such as variety, vigour, blossom development and weather conditions may influence the degree of success. Hoffman and Vangeluwe (1943) also sprayed a number of varieties with Elgetol, at blossom time, and found similar results. Elgetol showed more promise with some varieties than others. Flory and Moore (1944) found 0·2 per cent. concentration of Elgetol to be the most effective bloom spray for thinning York Imperial trees.

Schneider and Enzie (1943, 1944) also tested a number of materials for the purpose, e.g. liquid lime sulphur followed by summer oil, di-nitro compounds, Elgetol, Bordeaux mixture, Bordeaux and summer oil, naphthalene-acetic acid, naphthalene acetamide, indole-acetic acid and two commercial products known as Staymone and Hormex. These tests were carried out in various strengths and at various stages of flower opening. They came to the conclusion that liquid lime-sulphur followed by summer oil, Elgetol and Bordeaux spray combinations were not effective. Di-nitro compounds were effective but cresol damaged the trees slightly. Indole-acetic acid did not affect the crop but a spray of naphthalene-acetic acid, from 0·01 to 0·03 per cent. strength nearly eliminated the crop on the trees and damaged them severely which led them to suggest its application in the "off" year to reduce the fruit bud formation for the "on" year.

Murneek (1943) found that 1 to 2 per cent. of creosote oil and cresylic acid used in the "on" year completely destroyed the bloom on the trees of a number of varieties except Wealthy but there was little or no flower production in the following year. This led him to the conclusion that caustic sprays would prevent fruit set but that this factor alone was not sufficient to induce flower bud induction in the same year, especially when the trees were of relatively low vigour.

Gardner (1944) stated that several of the di-nitro-cresylic and phenolic compounds had two main drawbacks; firstly their effects varied from season to season, orchard to orchard, variety to variety, and even from tree to tree and, secondly, they often caused injury to the leaves and even to the bark and woody tissues of spurs. He thus sought to find materials which would reduce fruit set, not by killing the blossoms, but simply by coating over the stigmatic surfaces and thus preventing either pollination or pollen germination. He used oil wax emulsion (1 and 5 per cent.) at full bloom and reduced fruit set satisfactorily. This emulsion also acted as a sticker and insecticide and could reduce the rate of transpiration. Further Gardner, Stebbins and Neal (1946) reported three types of emulsions, which they named No. 82, No. 222 and T.S.S., to be most promising. They found that App-L set (sodium salt of naphthalene-acetic acid) and DN dry-mix No. 2, a di-nitro compound in powder form, also gave an effective result but suggested that App-L set (10 p.p.m.) plus oil wax emulsion No. 222 (1/400) may be used successfully for moderate thinning. They remarked, however, that the varieties differed greatly in the amount of thinning caused in them by the above emulsions.

Pruning. Crow (1920) suggested moderate heading back of small branches in the " off " year of Oldenburg and Wealthy varieties of apple, which would stop them from cropping heavily and thus help in correcting their biennial bearing tendency. Auchter and Schrader (1923) conducted experiments by giving special pruning to biennial bearing trees to stimulate spur growth and long terminal growth in the " off " year, in the hope of obtaining over-vegetative spurs which would not form blossom buds. Although they did not entirely succeed in their aim they remark that in conjunction with other cultural practices such as fruit thinning and manuring, pruning may help to overcome the alternate cropping habit. Roberts (1925) stated that pruning in the " off " year could induce annual bearing. Read (1931) also found " off " year spur pruning conducive to annual bearing. Thompson (1940) also stated that spur reduction before the " off " year, late in the season when blossom buds are visible, would help to correct biennial bearing. His idea was based on the principle that enough new wood should be available in a tree each season for the production of leaf surface and the development of blossom bud. He concluded, however, that this spur thinning would be most effective only if the tree had many weak and complex spurs.

Nitrogen manuring. Roberts (1920) suggested that early application of nitrogen in the spring of the " off " year may reduce the tendency of trees towards biennial bearing on the principle that nitrogen encourages the growth of spurs, some of which will thus become too vegetative to set fruit. Maney and Plagge (1925) also stated that nitrate manures encourage the growth in fruit spurs and vary the length of spurs and so affect the bearing of crop directly. Crow and Eidt (1921) recommended the early application of sodium nitrate in the spring of the " off " year to bring about regular bearing in biennial varieties. Hooker (1920), however, found that applying nitrogen in the late summer or autumn, led to more starch and more nitrogen in the tissue at the time of fruit bud differentiation in the following year. Carne (1931), Remy (1932) and McCormic (1933) have all suggested that nitrogen manuring, in general, may reduce the tendency of trees towards biennial bearing by maintaining appropriate vigour.

Dickson (1939) tried to influence biennial bearing by cultural treatments, especially by the application of manure and reported little response. In a further trial (1946), however, he found that the addition of farm manure for several years had to some extent modified the biennial tendency of a Baldwin orchard and suggested that the application of sufficient quantities of farm manure may partially correct biennial bearing.

Defoliation. Magness (1917) and Roberts (1923) claimed to have prevented the development of flower parts in the bud of the non-bearing spurs by removing their leaves. Harvey and Murneek (1921) and Haller and Magness (1933) also showed that there was considerable individuality in the buds and spurs and that they depended largely, if not entirely, upon their own leaves for development and blossom bud production. On the other hand Hooker and Bradford (1921) found with bearing trees that under some conditions there seemed to be no such localization of factors.

Harley *et al.* (1942) concluded from a number of defoliation and leaf/fruit adjustment studies that there was some specific factor responsible for the formation of flower primordia, probably a product of leaf metabolism. They did not know what particular substance constituted this factor and thought it might resemble a hormone substance.

They reached three important conclusions. Firstly, that there is a quantitative relationship between leaf area and blossom bud development ; secondly that the movement of the flower-producing substance is away from the bud until the requirements of other tissues have been met ; and thirdly that the active amount of leaf area needed to form the flower-producing substance appears to vary with the variety. In support of the statement that the flower producing substance moves to the roots they quoted the work of Davis (1931) in which he found that the roots of non-bearing prune trees showed a much higher percentage of starch than the non-bearing spurs.

Conservation of soil moisture at critical times. Many people are familiar with the widely prevalent idea that dry weather in spring helps to increase fruit bud formation. Sorauer, quoted by Gourley (1915), considered that decrease in supply of water and nitrogen salts and increase in phosphates and light were the essential conditions for fruit bud formation. Lees (1926) also put forward strong evidence in favour of a dry summer tending to increase fruit bud formation. Magness *et al.* (1932) found decreased carbohydrate formation in the leaves of trees in a dry summer but, on the contrary, an increased sugar/starch ratio in the plant. They thought that soluble sugar probably represented the more effective carbohydrate for the formation of fruit buds and this explained how drought conditions favoured fruit bud formation.

Degman, Furr and Magness (1933) also made detailed investigations of the relation of soil moisture to fruit bud formation of Oldenburg and Rome Beauty apple and found no correlation in the former ; but the trees of the latter that became dry formed a greater number of fruit buds.

Ringing. Ringing has been suggested as yet another way to exercise control over the formation of fruit buds. In this way Murneek (1939, 1940), Howlett (1941), Griggs and Schrader (1941) and many other investigators have induced fruit bud formation on branches and shoots which would otherwise have remained vegetative and unfruitful. It is believed that the slowing down of elongation growth and accumulation of starch as a direct outcome of ringing aids flower bud formation in some way. Ringing in this sense, however, is but a temporary measure and has very limited application.

Murneek (1943) studied the effect of branch ringing of York and Golden Delicious apple trees together with the use of chemical sprays normally employed to destroy blossoms. From his experiments he concluded that branch ringing of " on " year trees performed two weeks after destroying the flowers by means of sprays resulted in the production of more fruit buds than when sprays were used alone. It seems, therefore, that ringing carried out in conjunction with the use of chemical sprays provides a more efficient method of controlling biennial bearing than when either treatment is employed alone.

CONCLUSIONS

It is clear from the above review that the serious problem of biennial bearing still remains largely unsolved. The study of these past investigations indicates the complexity of the problem and makes it clear that no comprehensive remedy is likely to appear until the fundamental causes are more fully understood.

Previous studies have generally tended to emphasize single aspects, e.g. tree vigour, spur growth or chemical composition of spurs from " on " and " off " year trees. Alone these cannot give a clear picture of the whole phenomenon.

The problem is further complicated by inter-action and inter-dependence of various factors, e.g. a change in spur composition may be reflected in spur growth and vice versa. Similarly spur growth may be to some extent dependent on tree vigour and a change in tree vigour would automatically influence the spur growth. In addition these growth characters may vary to some extent according to climatic and environmental conditions which may further modify the final result. It is therefore apparent that in order to get a complete and real picture of the problem a comprehensive study of all the growth characters under the same environmental conditions is needed.

Similarly many attempts have been made to control biennial bearing by various treatments but in many cases workers have had conflicting results, e.g. some investigators have reported positive results with fruit thinning, others negative. The same is true of manuring and defoliation experiments. Of all the methods tried, blossom thinning seems to have yielded the most consistent results so far.

It appears to the author that biennial bearing is not the result of any one single factor, e.g. character of spur growth, amount of tree vigour, C/N ratio, or hormone activity, but that several such factors may be involved. In the series of studies which follow an attempt is therefore made to draw a picture of some of the important aspects of the problem on the basis of knowledge obtained from previous work. The new studies have been carried out in two main parts ; the first dealing with detailed growth studies in " on " and " off " year trees and the second with trials of some possible control measures. Reports of these studies are now ready for publication and should appear shortly.

ACKNOWLEDGMENTS.

The author expresses his warm thanks to Dr. W. S. Rogers who supervised this study, to Mr. D. Akenhead and other members of the Commonwealth Bureau of Horticulture and Plantation Crops and to Miss M. H. E. Davies, for help in the collection of references and in the presentation of this Review.

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(*Received* 27/2/48.)

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THE JONES-BATEMAN CUP FOR RESEARCH IN FRUIT GROWING.

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Vincent Square,
Westminster, S.W.1.

C. V. L. LYCETT, *Secretary.*

REVIEWS

LAND CLASSIFICATION IN THE WEST MIDLAND REGION. A Publication of the West Midland Group on Post-War Reconstruction and Planning. (London: Faber & Faber, 1947. Pp. 48. Price 12s. 6d.)

At a time when the face of England is threatened with or promised (according to one's personal viewpoint) a "planned" reorganization directed from Whitehall, a publication such as this brings hope that the counsel of those who have no political axe to grind and who have only the welfare of the country at heart may have a hearing. The publication of the Montague Barlow Report on the Geographical Distribution of the Industrial Population had at least one wholly beneficial sequel; a group of individuals in the West Midlands came together voluntarily to undertake a survey of the conditions within the region upon which any sound planning system must be based. Their first major publication was entitled "English County" and was a study of the regional planning problem in Herefordshire.

The approach to the problem in general is through a study of the land, its potentialities and actual use, and early in the work the difficulties of reconciling the outlook of the geographer and of the soil scientist became apparent. A compromise was finally reached and a band of workers within the Group made a broad survey of the whole Region on the basis of their agreed classification. The present work sets forth the results of the survey.

The scheme of classification adopted takes into account only factors of site and soil. In the first, relief, regional drainage and climate are the chief sub-sections, while the soil is considered from the viewpoint of its physical and chemical properties, more particularly the former. Part I of the book explains this classification in detail, while Part II describes the geographic and climatic features of the Region which provide the background for the survey. Parts III and IV constitute the details of the survey and the classification pattern of the Region. The last part presents a summary and the conclusions of the Group regarding the soil factors which should be taken into account in any future planning of the Region. Throughout, the report is illustrated with a series of sketch maps of admirable clarity and three large maps are provided showing the relief and geography, the geology and the land classification as a whole.

An interesting point to the horticulturist is that many of the most intensively cultivated areas, such as the Vale of Evesham and the Stourport region are on what is not by any means the best land. Other factors of site and human geography have been operative in the development of these areas.

If any criticism of so admirable a document is fair, one could regret that the conclusions drawn from the survey are so cautious and comparatively meagre. More forceful and categorical recommendations to guide the planners of the future might have been made, even if the likelihood of their adoption would appear to be slight.

The format, printing and paper of the book are of pre-war quality and make it a delight to read. As a contribution to the planning problem, throwing into relief the importance of the agricultural approach, the report is of the greatest value and it is to be hoped that similar surveys may be carried out for other regions.

R.H.S.

SEED PRODUCTION OF EUROPEAN VEGETABLES IN THE TROPICS.

By A. G. G. Hill. Technical Communication No. 19, Commonwealth Bureau of Horticulture and Plantation Crops (East Malling : Comm. Bur. Hort. and Plantation Crops. 1948. Pp. 28. Price 2s. net.)

Temperate region vegetables are grown in most tropical countries chiefly by or for European residents and are seldom crops of much importance in the native economy. Yet there are districts (for instance, in the West Indies) where they have attained a fairly extensive cultivation on a market garden scale and the total value of seeds imported annually from seedsmen in temperate regions amounts to a respectable figure.

The tropical zone embraces a wide range of climates and, although those of the lowlands are usually unfavourable for the flowering and fruiting of temperate species, those of the highlands may be quite suitable for seed production if the right methods are used. Even before the war, in the upland parts of East Africa, interest had been shown in local production of vegetable seeds and the Imperial Bureau of Horticulture and Plantation Crops had been asked to enquire into methods. When the war cut off many normal sources of seed supply, the interest was strengthened because it was realized that the tropical highlands might be able to produce seeds not only for tropical gardens but even for the United Kingdom and allied countries.

In October, 1944, the Bureau issued a questionnaire with the object of eliciting useful information from different parts of the tropics, and the Communication under review presents a summary of the replies received. Its present value, now that imported seed is again available in tropical countries, arises from the fact that the questionnaire was well framed to collect knowledge with a usefulness beyond the immediate emergency now happily passed.

In post-war colonial development more attention than ever before is being devoted to the nutrition of native peoples and increased green vegetable consumption is advocated and encouraged. Policy calls for more and cheaper production, and it is still an undecided question whether the more profitable line of attack is by genetic improvement of indigenous plants already used as vegetables or by breeding new varieties, better adapted to tropical conditions, of the temperate region species. Doubtless both lines offer promise and must be explored. For progress along either, improved techniques of seed production will be necessary and the information collated by the Bureau will find its application.

There is a bibliography of 41 references and useful appendices give lists of temperate region vegetable varieties grown in various tropical countries and of indigenous and naturalized plants used as vegetables.

E.E.C.

GEORGE HERBERT PETHYBRIDGE

LITTLE was it realized, when the last volume of this Journal was dedicated to Dr. Pethybridge on his relinquishing the post of Assistant Editor, that we should so soon have the melancholy task of recording his death. Although it is nearly twelve years since he retired from his official post in the Ministry of Agriculture and Fisheries, and seven since he went to live at his home town in Cornwall, it was with a real sense of loss that his wide circle of friends learned of his decease. He had a vivid personality, the memory of which is not easily dimmed by the passage of time.

Dr. Pethybridge was born at Bodmin, Cornwall, in 1871, his father and grandfather being well known in the district as managers of the old East Cornwall Bank. He was the seventh son in a family of twelve. After school days in Bodmin and at Dunheved College, Launceston, he followed the tradition of the family and entered a bank, but soon decided he did not like it. He then went to a Manchester school and matriculated through Owens College.

He began his scientific studies at the University College of Wales, Aberystwyth, where he was a contemporary of Sir John Russell, and continued them at the University of London, taking an honours degree in botany and geology. For a short time he taught science at Kingswood School near Bath, and then went to Germany to study at the University of Göttingen, where he obtained the degrees of M.A. and Ph.D. for research on the action of salts on plant tissues.

Shortly after his return from Germany he accepted a post at the Royal College of Science in Dublin, in a department which in 1900 was taken over by the newly created Department of Agriculture and Technical Instruction for Ireland. He soon began publishing papers on botanical and plant pathological subjects, one of his first, in the *Irish Naturalist* in 1903, being on " The Leaf-Spots of *Arum maculatum* ". In 1908 he was appointed Economic Botanist to the Department and at the same time Head of the Seeds and Plant Division.

It soon became evident that his main interest was in potato diseases. He felt that to gain the fullest understanding of these diseases he must take his microscope out into the field and study what was happening under natural conditions. Accordingly in 1909 he arranged for a small field laboratory and some ground for plots at Clifden in County Galway, on the west coast of Ireland. Here he was in the centre of a potato district and had only to go outside the door to find all the material for study that he wished. In this isolated spot he could work entirely free from interruption and he spent many weeks there each season, often with his assistant, Paul Murphy, who was later to make so many contributions of his own to the pathology of the potato. It was in this field laboratory that much of the careful observational and experimental work was done which appeared each year from 1910 to 1919 in the well-known series of reports, under the general title " Investigations on Potato Diseases ", in the Journal of the Department of Agriculture and Technical Instruction.

The reader of these reports was able to recognize at once that here was a scientist who saw the importance of studying the practical problems of the grower at their source—on the farm. And because these problems were so lucidly discussed in his annual reports and papers, a feeling of complete confidence in the work was engendered. According to a contemporary: "His reports to the Irish Department were to me the equivalent of the Bible in the potato world and I looked forward each year with eager anticipation to the appearance of the next."

It was also largely as a result of work done in the field laboratory at Clifden that the many carefully worked out papers on single diseases such as Dry Rot, Blight, Powdery Scab, Pink Rot, Verticillium Disease and the bacterial disease now known as Blackleg, appeared. Most of them were published in the Scientific Proceedings of the Royal Dublin Society or in the Proceedings of the Royal Irish Academy. References to them are included in a bibliography of thirty-eight titles accompanying a short appreciation of Pethybridge's work on potato diseases in No. 20 of Vol. XVI (N.S.) of the Royal Dublin Society's Proceedings for 1921, on the occasion of the award to him of the Boyle Medal. This paper describes briefly his investigations into the life-cycle of the Potato Blight fungus and his conclusion that oospores play no part in it in nature, and also his discovery of a new species of Phytophthora (*P. erythroseptica*) with a most interesting new antheridial-oogonial relationship.

Although potato diseases were Dr. Pethybridge's chief interest, they were by no means his only one. In the years before 1910 he had collaborated in papers entitled "The Vegetation of the District lying South of Dublin" and "A Census Catalogue of Irish Fungi". As Economic Botanist to the Department he received numerous inquiries on seed-borne diseases, vegetable diseases and other subjects. During the first world war the production of flax for aeroplane manufacture became of first-class importance. Very little was then known about flax diseases, however, and in 1917 he turned his attention to studying them. Profiting by the experience gained in the use of the field laboratory for investigations on potato diseases, he decided to pursue a similar course in respect of flax diseases and had a special laboratory erected for the purpose near Coleraine, Co. Derry. Detailed papers on flax diseases, similar to those on the potato diseases, soon began to appear as a result of this initiative, his collaborator in this work being H. A. Lafferty.

On all the problems Pethybridge touched he made exact observations and useful contributions to knowledge. He refused to be submerged by administrative work and insisted on finding out for himself, as far as possible, the answers to all problems referred to his laboratory, however small.

He was imbued with the keen enthusiasm for discovery which characterized most students of biology in the latter half of the nineteenth century, when new horizons were being opened up by the microscope and fresh fields charted by the methods of exact experiment. It is not easy for us of a younger generation to realize the sense of adventure inherent in the carrying out of research in biology in those days. The scientific world was smaller; one knew the work of one's contemporaries intimately; not infrequently one found oneself discovering new facts missed by them, or disputing with them about interpretations. Something of this "atmosphere of discovery" is revealed in the manner in which the results of work were recorded in scientific journals of the day. Most minute descriptions of the facts observed and of the steps taken in reaching conclusions were often given. This style of writing was exactly suited to Pethybridge's critical temperament and his papers were models of lucidity for the period in which he wrote.

With the change of Government in Ireland in 1923, Dr. Pethybridge resigned his position and shortly afterwards accepted the post of Mycologist to the Ministry of Agriculture and Fisheries at the Plant Pathology Laboratory, Harpenden. Here his duties consisted not merely in advising the Ministry on technical matters concerning plant diseases, but also in collating records that came in to the Laboratory on the prevalence of diseases, and in acting as Chairman of periodical meetings of the Advisory Mycologists in the Agricultural Advisory Service. He was later made Assistant Director of the Laboratory.

His continued interest in the details of plant diseases is evident from the numerous special notes included in his Reports on Fungus, Bacterial and Other Diseases of Crops in England and Wales (e.g. in Bull. 79 (1934) of the Ministry of Agriculture and Fisheries). Another effect of his coming to England was the stimulating influence he exerted on his new and wider circle of professional colleagues. All recognized in him a plant pathologist of exceptional practical experience, who demanded a high standard of accuracy in any work with which he was connected. To some he seemed at times a little super-critical, but this did not lessen the general esteem in which he was held. Many younger men, especially, are grateful to him, both for the help he gave them in their work and for advice on matters affecting their careers.

Dr. Pethybridge was a valued member of a number of professional societies, including the Royal Irish Academy, the Royal Dublin Society, the Linnean Society, the Royal Horticultural Society, the Association of Applied Biologists and the British Mycological Society, and at different times was elected to the Councils or held the Presidency or Vice-Presidency of several of them. He had a quiet humour and a fund of stories which enlivened many of the more informal gatherings of plant pathologists.

In 1936 he retired from the Ministry of Agriculture and Fisheries and the following year his services were recognized by his appointment as Officer of the British Empire (Civil Division).

As already mentioned in the Editorial to Parts 3 and 4 of Vol. XXIII of this *Journal*, issued last December, the Joint Editors secured his services as Assistant Editor on his retirement from his official posts in 1936. For nearly eleven years he gave unstinted service to the *Journal*, devoting much time to that often thankless task of editors—suggesting to authors ways of improving the presentation of their papers. Undoubtedly it was a shock to some to receive their manuscripts back with proposals for drastic curtailment and with many corrections in the English; but there are few who would not have admitted a year or two later that they had learnt valuable lessons as a result of their experience.

Even in his retirement at Bodmin he continued to conduct an extensive correspondence with plant pathologist friends of former days, welcoming any who travelled that way, attending wartime demonstrations of potato spraying, and occasionally even giving demonstrations himself at the local school. Right up to the last his microscope was in use to satisfy his own curiosity or to help or instruct his friends.

After a short illness he died on May 23rd, 1948. There can be no plant pathologist who knew him who did not learn with regret of the passing of " Pethy ".

G.S.

THE APPLICATION OF RAPID CHEMICAL TESTS TO THE DIAGNOSIS OF MINERAL DEFICIENCIES IN HORTICULTURAL CROPS

I. CROPS GROWN ON A MANURIAL TRIAL

By **D. J. D. NICHOLAS**
Long Ashton Research Station, Bristol

SYNOPSIS

A chemical tissue test method, designed to determine the mineral status of field crops, which has been tested to diagnose mineral deficiencies in potato and cauliflower, is described. The procedure comprises a specialized method of sampling fresh plant material, followed by the extraction of the easily soluble nutrients from selected portions of the plants and their subsequent estimation by colorimetric and turbidimetric tests. A buffered solution consisting of glacial acetic acid and sodium acetate (100 gm. sodium acetate and 30 ml. of glacial acetic acid in 1 litre of distilled water, at pH 4·8) is used for the extraction of the nutrients from weighed amounts of fresh tissue for a standard time, viz. 15 minutes. The tissue extract is tested for potassium, magnesium, calcium, phosphorus, nitrate nitrogen and manganese. The detection of the last at deficiency levels involves the use of specially purified reagents. Nutrient standards for the various elements have been fixed for a number of plants and threshold deficiency values are given which coincide with the development of visual symptoms of the respective deficiencies. It is shown that the tissue test data reflect manurial and cultural treatments and are in accord with results of the visual method and those of ash analysis of the plants. The method is recommended as complementary to visual diagnosis for determining deficiencies in field crops and as a quick procedure for assessing the mineral status of plants. It replaces the time-consuming process of ash analysis.

THE practice of determining the mineral requirements of crop plants by means of chemical tissue tests originated in America. Early work was done by Hoffer (22) who made qualitative tests for Fe and K in fresh maize stalks (*Zea mays*) and showed that Fe accumulated at the nodes. He noted an inverse relation between K and Fe status of the nodal tissues and used this staining method to confirm K deficiency. McGillivray *et al.* (25), using thin cross-sections of leaf petioles of tomatoes immersed in platinum chloride solution for twenty-four hours, made microscopic studies of the relative abundance of potassium platinic chloride crystals. This was used as an indication of the K status of the plants.

A later development was the study by Gilbert and Hardin (18) of the nutrient content of expressed sap in regard to N, P and K, by grinding fresh material and straining through a muslin mesh. McCool and Weldon (24) used a similar method for K and P using sap extracted at a pressure of ½ to 1 ton per sq. in. Fonder (14, 15) and Cook (6, 7) examined the sap of beans and cereals respectively for Ca and Mg and nitrate N. Pettinger (40) showed that the sap of maize reflected the N, P, K treatments given and in some instances Cl was also included. Neller (31) determined P in plant sap of sorghum and buckwheat, and Poehlman (43) nitrate N, P, K; both showed that it was impossible to fix critical levels for the nutrients as had been recommended by Carolus working with potatoes (3, 4). Discrepancies in the composition of extracted sap have been noted by Knudson and Ginsburg (23) and Gassner and

Goeze (16, 17) especially when the pressure used to obtain it is varied or when there is a time difference in attaining the same trial pressure. Phillis and Mason (41) confirmed these variations using fresh plant tissue and it was suggested that freezing of tissues or ether treatment might be necessary to destroy the semi-permeable membranes of the cells. Work along these lines has not yielded consistent results and no publications have appeared in recent years.

The extraction of soluble nutrients has been attempted by a number of workers, using a variety of solvents. The simplest procedure is the extraction with water as used by Nightingale (36, 37) for the detection of Ca deficiency in sugar cane and pineapple, and by Marsh (27) using tobacco and maize. Page and Burkhart (38) have used boiling water for the extraction of nutrients from fresh material for diagnosis of NPK and deficiencies in peanut and cotton. The use of an organic acid, 2 per cent. acetic acid, for extraction, was introduced by Emmert (8, 9, 10, 11, 12, 13) for detecting nitrate N, P and K, and later Carolus (3, 4) extended it to include Ca and Mg in vegetable crops. These investigators as well as Hill (21), Cook (6, 7) and Ulrich (47) have reported satisfactory results but New Zealand work reported by McNaught (26) does not substantiate these claims.

The extraction of nutrients by the Purdue method devised by Thornton *et al.* (45, 46) is made by the chemical reagents used for detecting the required nutrient, e.g. cobaltinitrite for K, ammonium molybdate for P, and diphenylamine in sulphuric acid for nitrate N. Tests, which have been made on maize and cotton, are only roughly quantitative because of the fact that the tissue is *in situ* and nutrients continue to diffuse out into solution. The results are usually reported in four categories, viz. very high, high, medium and low, but numerical data for these categories have not been reported. This method has been used by Scarseth (44) Atkinson (2) and Wark (51) for the diagnosis of mineral deficiencies in crop plants.

Carolus (3, 4), Hester (20) and Peech and English (39) have used a Waring blendor for extracting the nutrients from fresh tissues. Carolus used a 2 per cent. acetic acid but Hester adopted the acetate buffer solution (pH 4·8) as developed by Morgan (28, 29, 30) for the latter's soil testing system. Hester has suggested values for nitrate N, P and K which correspond to maximum yield data in vegetable crops. These methods, however, have the disadvantage of not being usable in the field.

A tissue test system has been developed at Long Ashton (33, 34, 35, 42) which is comparable to Thornton's method for speed. It includes a more precise quantitative determination of the soluble nutrients and is capable of use in the field.

This paper describes the application of the method in the study of the mineral status of potato (vars. Majestic and Kerr's Pink) and cauliflower grown on a long term manurial trial. It is shown that the correlation between the tissue test data and results of other methods used to determine mineral status, in particular those of the visual method (49) and ash analysis of the foliage, is usually close.

EXPERIMENTAL.

Materials.—Table I contains data concerning the site, soil characters, crops grown, and nutritional problems occurring at the centre, as well as the sampling times for tissue tests and ash analysis.

At this centre use was made of a long term manurial trial in which six treatments are represented: Nil, farmyard manure, complete fertilizer (NPK), Omit nitrogen

TABLE I.

Centre.	Sampling dates for tests and ash analysis.		Soil characters.	Crop and variety.	Nutritional problems.
A. Long Ashton Plot 17A	2/7/45 25/7/45 23/8/45	12/6/46 8/7/46 20/8/46	Calcareous sandy loam derived from red drift overlying Keuper Marl.	Potato, Majestic and Kerr's Pink	N, P (trace), K and Mg deficiencies.
	17/7/45 27/8/45 21/9/45	29/7/46 29/8/46 25/9/46		Cauliflower, Majestic	N, K and Mg deficiencies

(PK), Omit phosphorus (NK), Omit potassium (NP). The annual rates of application per acre were as follows :

F.Y.M.	10 tons per acre.
Nitrogen	4 cwt. of nitrochalk per acre.
Phosphorus	3 cwt. of superphosphate per acre.
Potassium	2 cwt. of sulphate of potash per acre.

The treatments occur in triplicate and magnesium sulphate at 1 cwt. per acre is applied annually to one set of the six treatments (Block II).

The surface soil and subsoil contain approximately 5 per cent. and 1 per cent. free lime respectively with a pH of about 7·2 to 7·4 in the top soil.

METHODS FOR CHEMICAL TISSUE TESTS AND ASH ANALYSIS.

Selection of plant parts.

Details of the field sampling method have been given in previous papers (33, 34, 35, 42). The importance of taking morphologically similar parts of plants for comparison has been stressed in view of the presence of nutrient gradients within plants. The leaves taken for examination are as follows :

Potato and tomato : Leaves from the mid-stem portion of plants.

Cauliflower : The first upright leaf outside incurled leaves.

Apple and black currant : The 3rd and 4th basal leaves taken from well exposed leader shoots.

Preparation of test samples.

(*a*) *Tissue tests.* Small petiole portions taken from numerous leaves at approximately half-way between leaf attachment and base of lamina are used. In the cauliflower, mid-rib portions of comparable girth near the tip of the lamina are usually taken.

(*b*) *Ash analyses.* The leaves, including the petiole, are dried for 48 hours in a specially aerated oven and are then milled. When a wet digestion procedure is used for the removal of organic matter, approximately 5 gm. of dry matter is placed in a 40 ml. pyrex beaker fitted with a watch glass and 10 ml. of HNO_3 (conc.) added per 1 gm. This is heated on an Argand burner for approximately 35 minutes to remove ammonia. After cooling, 1 ml. of $HClO_4$ and 0·1 ml. H_2SO_4 per gm. of dry matter are added slowly and heating is continued until the " frothing " stage, when the oxidation of organic matter takes place. At this stage the oxidation process is continued at a lower temperature until the silica is dehydrated. Fifty ml. of H_2O is then added and further heated for 15 minutes, cooled and filtered through a 42 Whatman paper into a graduated flask.

Extraction of soluble nutrients

Morgan's reagent, consisting of 100 gm. of sodium acetate and 30 ml. of glacial acetic acid (pH 4·8) is used for the extraction of soluble K, Mg, Ca, P, nitrate N, Cl and Mn (for the last when present in excess). The method recommended in previous papers (33, 34) is used, viz. 4 gm. of finely chopped tissue is immersed in 40 ml. of Morgan's reagent for 15 minutes and the resultant solution is then poured off quickly into a clean specimen tube, using a glass funnel fitted with a small plug of cotton wool.

The tissue extract is usually clear, but those of fruit tree petioles may be coloured by flavones, tannins and other phenolic compounds. These may be removed before testing by using 0·2 gm. of specially purified carbon (Darco G.60 grade) per 25 ml. of extract and stirring with a glass rod for 30 seconds, then filtering through a No. 41 Whatman paper into another specimen tube. A purified carbon may be prepared by boiling the B.D.H. product with 5 N. HCl and rinsing with hot distilled water. The addition of sodium chloride facilitates the removal of phosphate from the carbon. Rinsing with hot water should continue until Cl cannot be detected in the leachate and then, after drying, the carbon is tested for mineral elements by shaking with Morgan's solution. The results obtained on blank determinants are usually <1 p.p.m. PO_4; 0·2 p.p.m. Mg; Nil K; Nil Ca.

For the detection of deficiency levels of Mn in crop plants, a purified Morgan's reagent is used (34). The last traces of Mn may be removed from sodium acetate by using a modification of Arnon and Stout's adsorption method (1) as follows: 100 gm. of sodium acetate are dissolved in 500 ml. of distilled water and autoclaved at a temperature of 120° C. for three-quarters of an hour in the presence of 20 gm. of $CaCO_3$ and 20 ml. 20 per cent. solution of Na_2HPO_4 and $CaCl_2$ respectively. The presence of 5 gm. each of Na_2CO_3 and $NaHCO_3$ helps to remove Mn completely. It has also been noted that should the Mn impurity be $<40 \times 10^{-3}$ p.p.m. Mn, the addition of 5 ml. of 25×10^{-3} p.p.m. to the autoclaved mixture assists in the complete removal of Mn from solution, i.e. $<1 \times 10^{-3}$ p.p.m. Mn.

Chemical tests used.

Since the initial progress report (32, 42) several modifications and refinements have been made. The chemical tests used are given in Table II.

The following notes are added to supplement the details in the table.

Potassium. The sodium cobaltinitrite should be freshly prepared every three weeks, thoroughly aerated before use and stored in the refrigerator. After 2 ml. of isopropyl alcohol is added, the tube is shaken vigorously, using a clockwise movement for one minute to effect a complete precipitation of the potassium sodium cobaltinitrite. The shaking procedure must be standardized for consistent results. The turbidity is determined immediately, using the Spekker Absorptiometer with red Ilford filters and a water setting of 1, using 1 cm. cells.

Calcium. After the addition of the oxalate the tube is shaken as for the K method for 2 minutes and is then allowed to stand for 15 minutes. The turbidity is determined on the Spekker, using blue filters with a water setting of 0.8, and 1 cm. cells. Care must be taken to transfer the total precipitate to the Spekker cells.

Magnesium. For most of the Long Ashton work before the war, Titan Yellow, (Dr. Grubler grade) has been used and this has proved most reliable. Titan Yellow however, may vary in strength and quality with batches and thus it is necessary to recrystallize and standardize the product. The latter may be conveniently done by making up the recrystallized material in alcohol (see Table II) and then adding 0·1, 0·2, ... 1·0 ml. respectively to a series of tubes containing 5 ml. of 5 p.p.m. Mg standards and completing the test. Matching is then done with a Lovibond disc and the volume of Titan Yellow required for perfect match is always used for that batch of the dye. Titan Yellow standards must be checked at frequent intervals.

TABLE II.

Chemical tests.

(*Using Morgan's solution as the extracting solution.*)

Kations or anions tested.	Reagents used.	Coloration or turbidity ranges.	
		Minimum.	Maximum.
Potassium (K) ..	0·2 ml. 35% sodium cobaltinitrite. 1 ml. 50% glycerin. 2 ml. isopropyl alcohol.	Clear reddish brown solution	Deep canary yellow turbidity.
Calcium (Ca) ..	2 ml. 50% glycerin. 5 ml. saturated solution of ammonium oxalate.	Colourless solution.	Greyish white turbidity.
Magnesium (Mg)	0·2 ml. 0·15% Titan yellow.* 0·5 ml. 2% hydroxylamine hydrochloride. 0·5 ml. 5% sucrose. 2 ml. 10% sodium hydroxide.	Straw yellow solution.	Salmon pink colour.
Manganese (Mn) (present in excess)	0·5 ml. 10% trioxymethylene sulphate. 2 ml. 10% sodium hydroxide.	Colourless solution.	Deep cherry colour.
Manganese (Mn) (sensitive test for deficiency levels)	2 ml. potassium periodate.* (Saturated solution.) 0·4 ml. 1% tetramethyl.*	Pale blue colour.	Deep blue colour.
Nitrate (NO_3) ..	2 ml. 25% W/V phenoldisulphonic acid in sulphuric acid. 5 ml. H_2O. 10 ml. 30% ammonia until alkaline.	Colourless.	Deep buff colour.
Phosphate (PO_4)	2 ml. 4% ammonium molybdate. 1 ml. 1% hydroquinone. 2 ml. {26% potassium carbonate. 5% sodium sulphite.	Faint blue colour.	Deep Mediterranean blue colour.
Chloride (Cl) ..	2 ml. N/50 silver nitrate. 3 drops HNO_3 (conc.).	Colourless solution.	White turbidity.

* Should be recrystallized and standardized before use.

TABLE III.

Relation between tissue test values and actual concentration of nutrients in parts per million in the Morgan's Extract (normal and deficiency levels).

	Potassium.		Calcium.			Magnesium.		Manganese.	Phosphate.		Nitrate.	
	Cauliflower, Tomato, Black currant.	Potato, Apple.	Tomato, Potato.	Apple.	Cauli-flower.	Apple.	Potato, Tomato, Cauliflower, Black currant.	Tetramethyl method. Potato, Cauliflower.	Potato, Tomato, Black currant, Apple.	Cauli-flower.	Potato.	Tomato, Cauli-flower.
High + ..			270				14	80×10^{-3}				350
High ..	200	150	200	175	300	8	12	70×10^{-3}	30	60	150	300
High − ..	165	125	165	150	250	7	10	60×10^{-3}	25	50	125	250
Medium +	130	100	130	125	200	6	8	50×10^{-3}	20	40	100	200
Medium ..	95	75	95	100	150	5	6	40×10^{-3}	15	30	75	150
Medium −	60	50	60	75	100	4	4	30×10^{-3}	10	20	50	100
Low + ..	25	25	25	50	50	3	2	20×10^{-3}	5	10	25	50
Low ..	10	10	10	10	25	2	1	10×10^{-3}	<5	<10	<10	<25
Low − ..	<10	<10	<10	<10	<25	1	<0·5	5×10^{-3}				

Manganese (*at deficiency levels*). Tetramethyldiaminodiphenylmethane should be recrystallized before use, using redistilled methyl alcohol. The tetrabase should be colourless when dissolved in acetone. The colour is more stable when the tissue extracts are cooled in ice.

Nitrate. Nitrates are usually determined by evaporating the solution to dryness and then adding phenoldisulphonic acid or brucine in H_2SO_4. Attempts were made initially to add phenoldisulphonic acid to macerated tissues but charring masked the yellow colour. It was found that the addition of phenoldisulphonic acid to the tissue extract, followed by ammonia after cooling, gave differences during the early growth period between normal plants and those deficient in nitrogen. Later in the season it was found that nitrate N fell to low values in tissue extracts, irrespective of treatment. and the evaporation of the tissue extract and addition of phenoldisulphonic acid to the residue did not produce a nitrate colour, thus proving the absence of nitrate N in plants late in the season.

Nitrates could not be detected in either apple petiole extracts or those of legumes even though the extracts were evaporated to dryness and phenoldisulphonic acid added to the residue.

RESULTS.

Nutrient standards.

The assessment of colour and turbidity is made by using a Lovibond Comparator and discs, or, for more precise work, by prepared standards made up in Morgan's solution calibrated by means of the Spekker Absorptiometer. An empirical scale of values is used for each nutrient comprising High (H), Medium (M), Low (L), and Trace (Tr), and these categories may be further divided by the use of a + or − sign. The values are fixed for each crop plant. Approximate relationships between tissue test categories and concentration of nutrients in the extracts of various crop plants are given in Tables III and IV.

TABLE IV.

Relation between visual tissue test groupings and concentration of Mn and Cl in parts per million in the Morgan's Extract (normal and toxic levels.)

Visual groupings.	High ++++	High +++	High ++	High +	High	High −	Trace ++ or medium.	Trace +	Trace
Mn	10	8	5	3	2	1	0·8	0·5	0·2
Cl	150	125	100	75	50	25	15	10	5

A series of colour standards using bromothymol blue, thymol blue and bromophenol blue are used in the determination of Mn by the tetrabase method. Matching of Mn in the tissue extract should be made within 1 minute of adding the reagents and not later than 2 minutes, as the reaction is catalytic and, at high Mn levels, the blue colour gives way to fluorescent red and green, making matching with colour standards difficult. Data relating to the making up of the colour standards are found in Table V. These standards were matched individually with Mn standards made up in the Morgan's reagent.

TABLE V.

Colour standards for the determination of manganese in tissue extracts (tetramethyl test) showing amounts of bromothymol blue, thymol blue, bromophenol blue and ammonia (volume in mls.).

	Lettered labels to facilitate quick matching.									
Indicator used.	A.	B.	C.	D.	E.	F.	G.	H.	I.	J.
Bromothymol blue ..	0·03	0·06	0·09	0·125	0·15	0·20	0·25	0·30	0·40	0·50
Thymol blue ..				0·025	0·05	0·10	0·10	0·15	0·15	0·20
Bromophenol blue ..							0·05	0·10	0·15	0·20
2% NH_4OH ..	13	13	13	13	13	15	18	18	18	20
Mn in p.p.m. $\times 10^{-3}$ ..	2–3	5	10	15	20	30	40	50	60	80

Tissue extracts must be chilled in an ice salt mixture for 5 minutes before testing.

At this centre, tissue tests for K, Mg, Ca, PO_4, nitrate N, and Mn were made at six fortnightly intervals throughout the 1945 and 1946 seasons using two varieties of potato (Majestic and Kerr's Pink) and cauliflower. Ash analyses of the Majestic variety and of the cauliflower were made three times during the seasons 1945 and 1946, coinciding with tissue test determinations. The data are presented as histograms.*

The results for both crops are considered under the following heads.

TISSUE TESTS IN RELATION TO MANURIAL AND CULTURAL TREATMENTS.

A series of histograms representing the mean of the six seasonal tissue test values for K, Mg, Ca, PO_4, nitrate N and Mn in potato (Majestic and Kerr's Pink) and in cauliflower for the seasons 1945 and 1946 are given in Figs. 1 to 5.

Potassium. There is a significant difference between treated and non-treated K plants and the NP (Omit K) levels are usually lower than in the Nil potato and cauliflower. The K values in potato, where K is omitted, are lower than in cauliflower on the corresponding plots, which is in agreement with the greater susceptibility of the former to K deficiency.

Magnesium. The Mg content of PK (Omit N) plants are lower than those of other treatments in Blocks I and III, and this is particularly marked in the latter. Block II plants reflect the magnesium sulphate soil treatment but the Omit N plants still show the lowest Mg value within the block. During the 1946 season the Mg level in the NPK plants (Blocks I and III) is also low, probably due to the wet season

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* Data in tabular form are available for consultation at Long Ashton.

SEASON 1945.

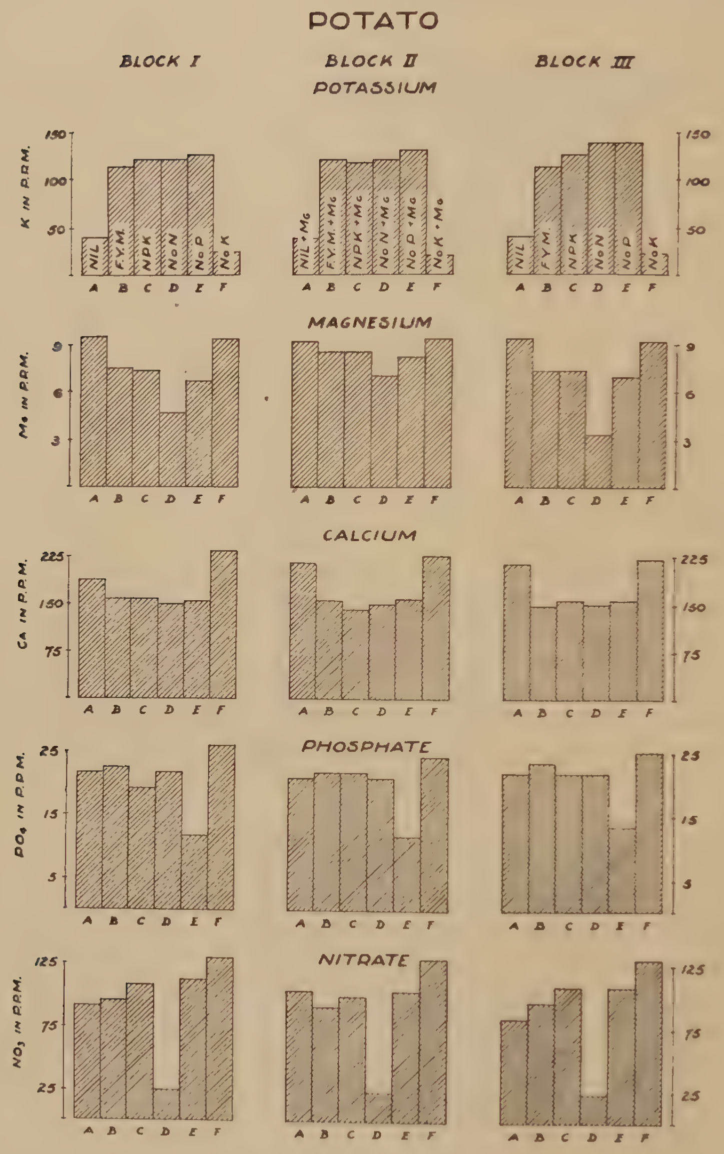

FIG. 1.

Histograms representing the mean of the six seasonal tissue test results for K, Mg, Ca, PO_4 and NO_3 in POTATO (Majestic) growing under various manurial treatments. Season 1945.

SEASON 1946.

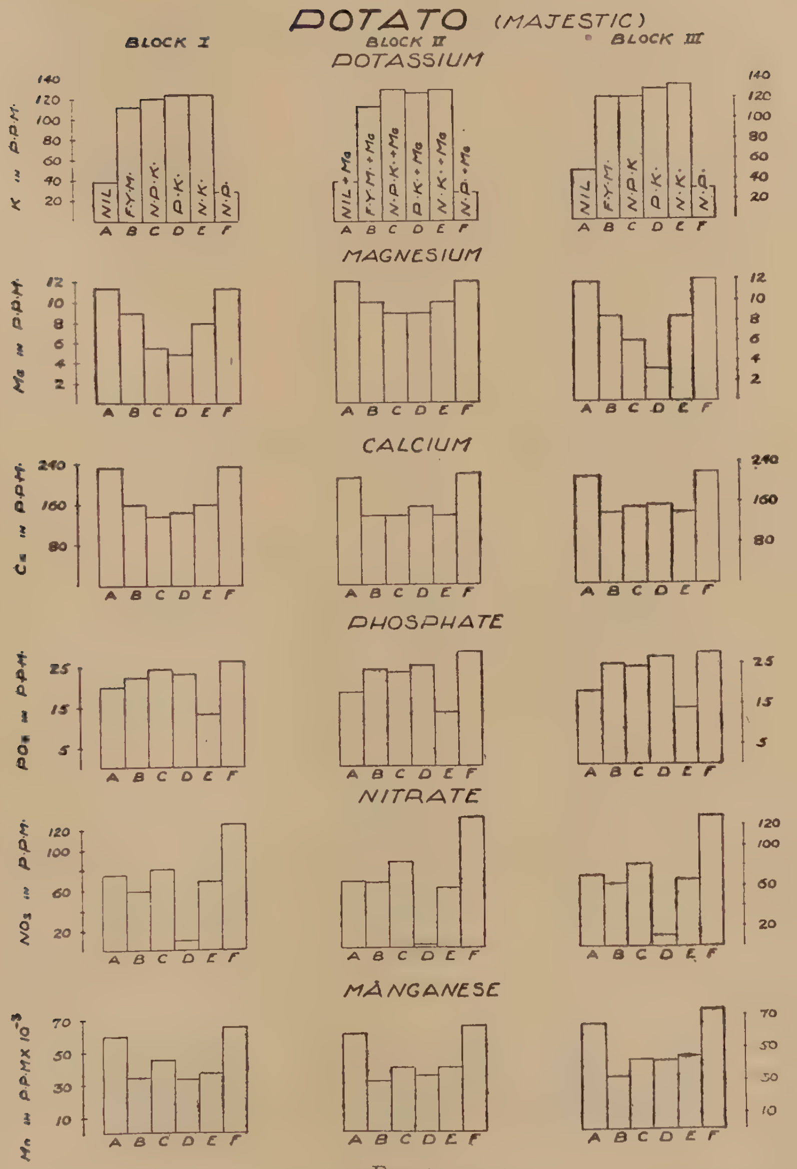

FIG. 2.

Histograms representing the mean of the six seasonal tissue test results for K, Mg, Ca, PO_4, NO_3 and Mn in POTATO (Majestic) growing under various manurial treatments. Season 1946.

SEASON 1946.

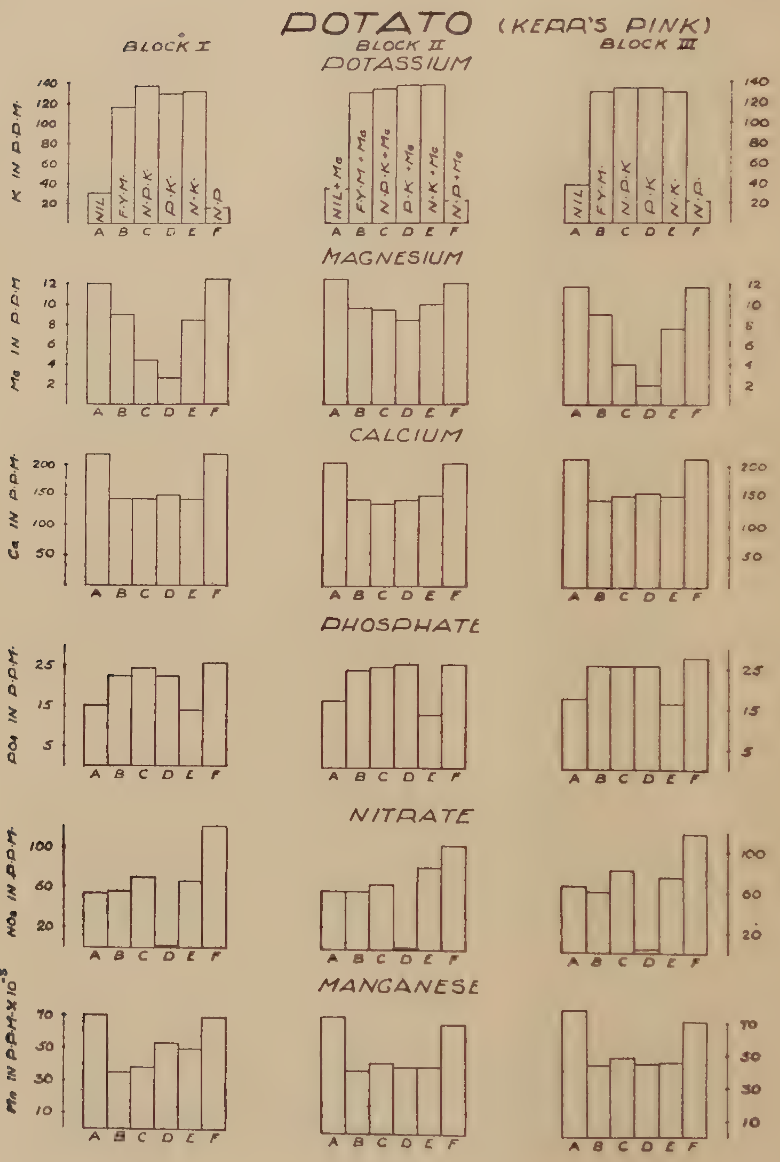

FIG. 3.

Histograms representing the mean of the six seasonal tissue test results for K, Mg, Ca, PO_4, NO_3 and Mn in POTATO (Kerr's Pink) growing under various manurial treatments. Season 1946.

SEASON 1945.

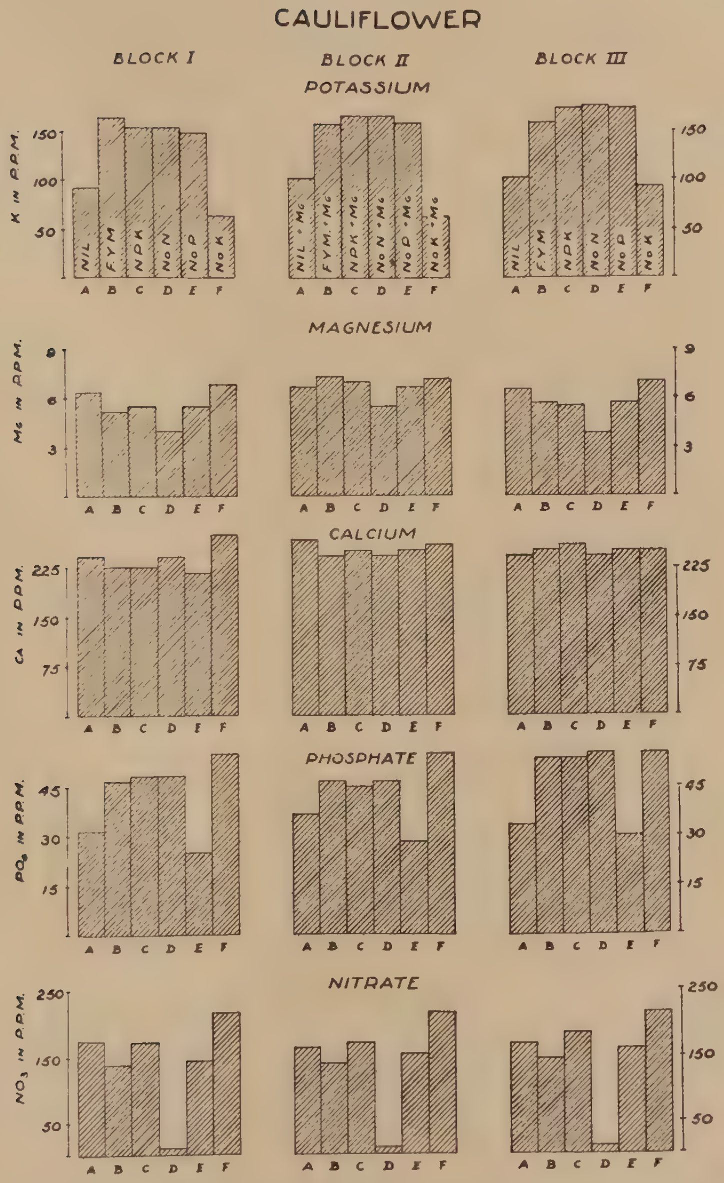

FIG. 4.

Histograms representing the mean of the six seasonal tissue test results for K, Mg, Ca, PO_4 and NO_3 in CAULIFLOWER growing under various manurial treatments. Season 1945.

SEASON 1946.

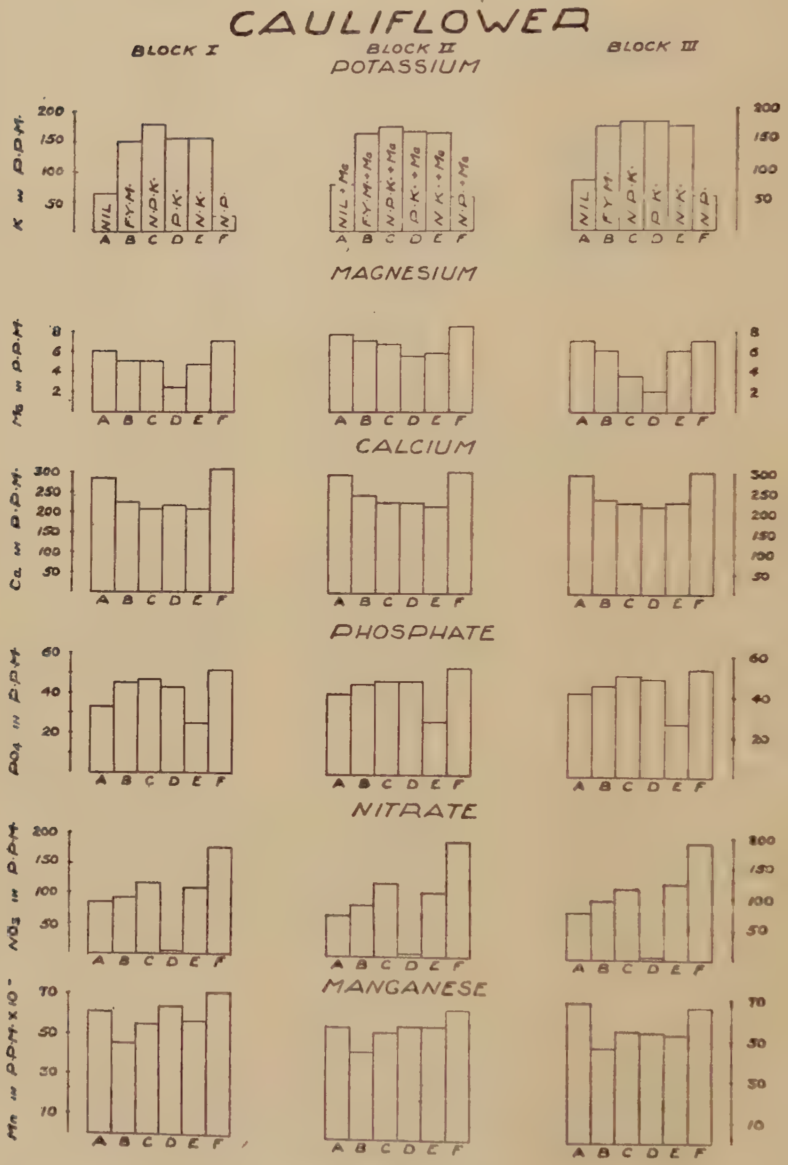

FIG. 5.

Histograms representing the mean of the six seasonal tissue test results for K, Mg, Ca, PO_4, NO_3 and Mn in CAULIFLOWER growing under various manurial treatments. Season 1946.

SEASON 1945.

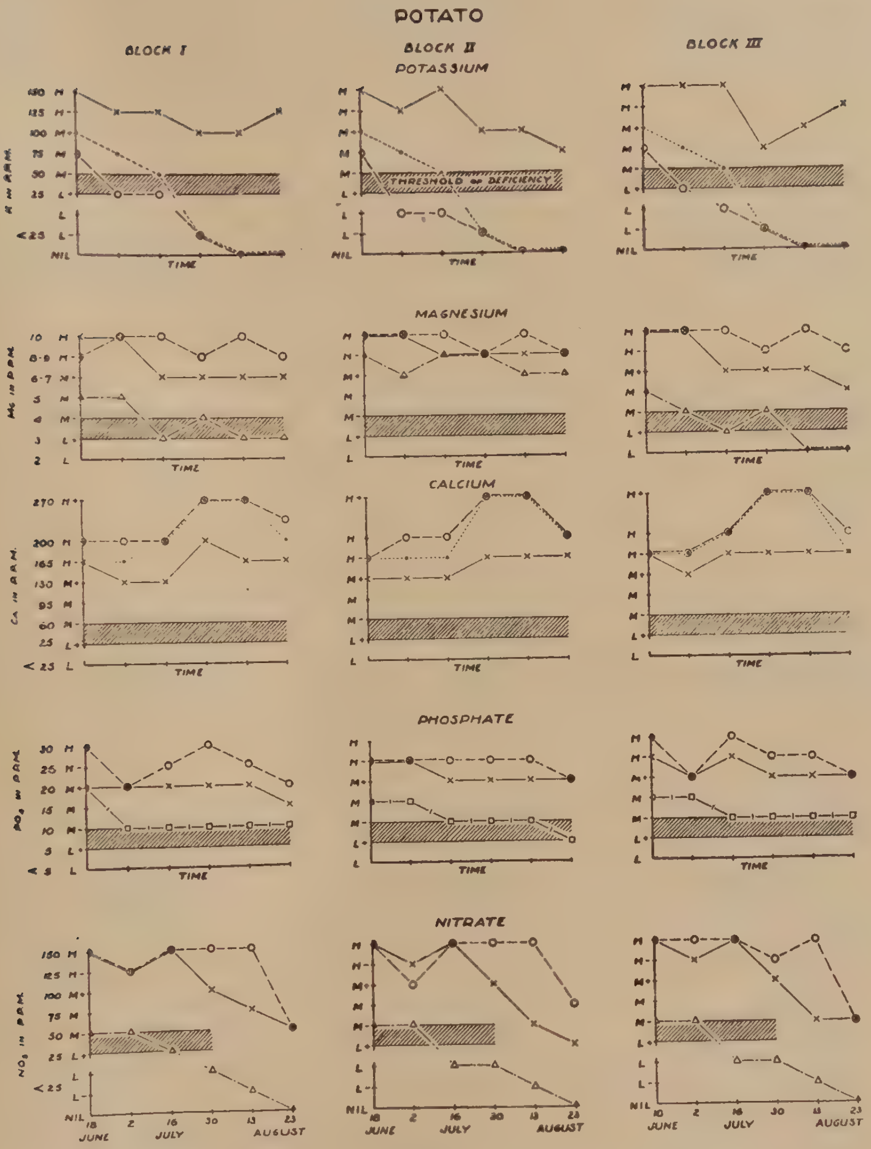

FIG. 6.

The Seasonal Cycles of soluble K, Mg, Ca, PO$_4$ and NO$_3$ in POTATO (Majestic).
The following treatments are represented :
(1) × ———— × NPK. (2) • . . . . • Nil. (3) ○ ———— ○ NP.
(4) △ —— – —— △ PK. (5) □——|——□ NK. Season 1945.
The shaded area represents the threshold range associated with visual signs of a deficiency.

SEASON 1946.

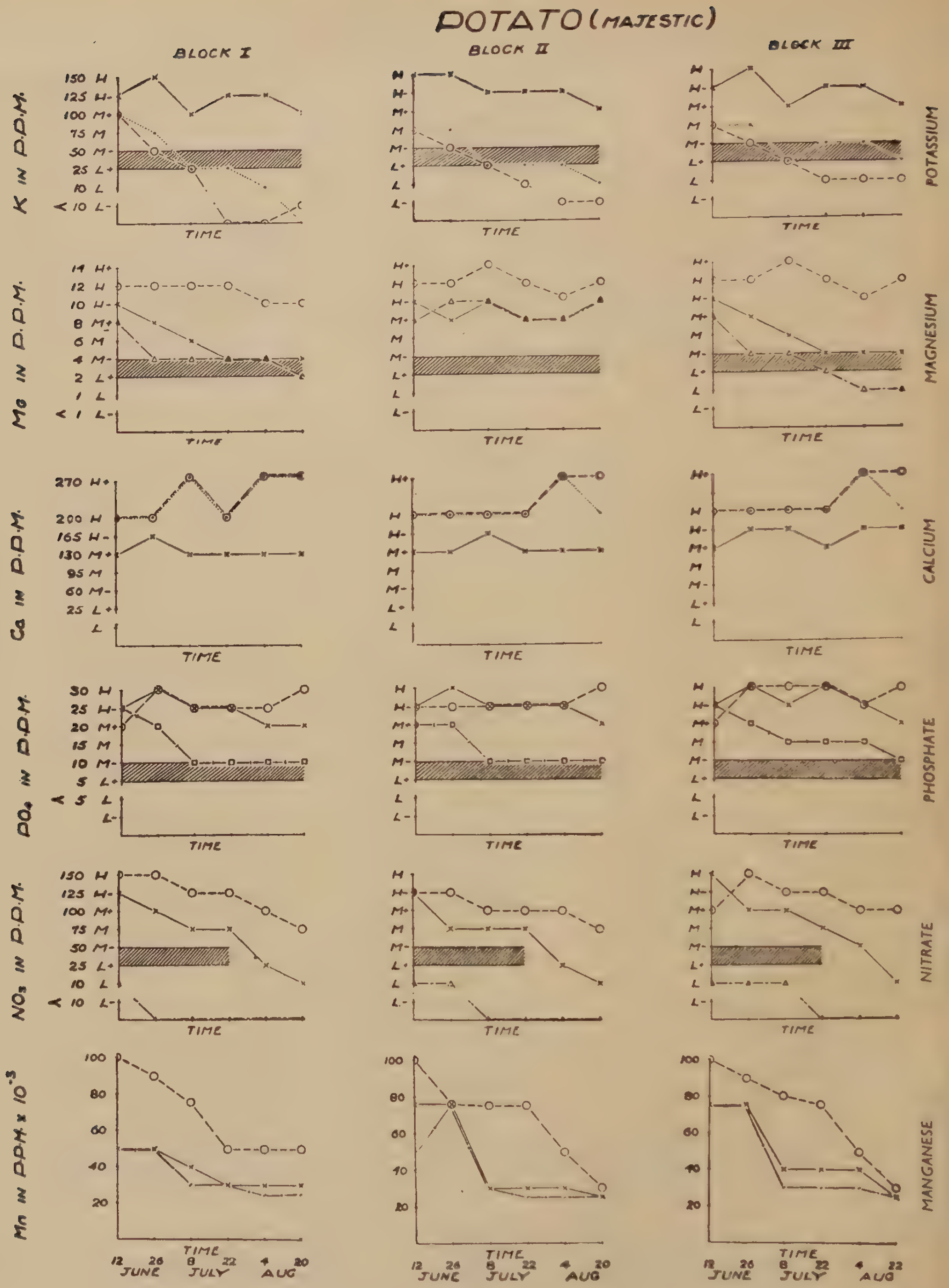

FIG. 7.

The Seasonal Cycles of soluble K, Mg, Ca, PO_4, NO_3 and Mn in POTATO (Majestic). The following treatments are represented:
(1) ×————× NPK. (2) •..........• Nil (3) ○————○ NP.
(4) △—·—·—△ PK. □——|——□ NK Season 1946.
The shaded area represents the threshold range associated with visual signs of a deficiency.

SEASON 1946.

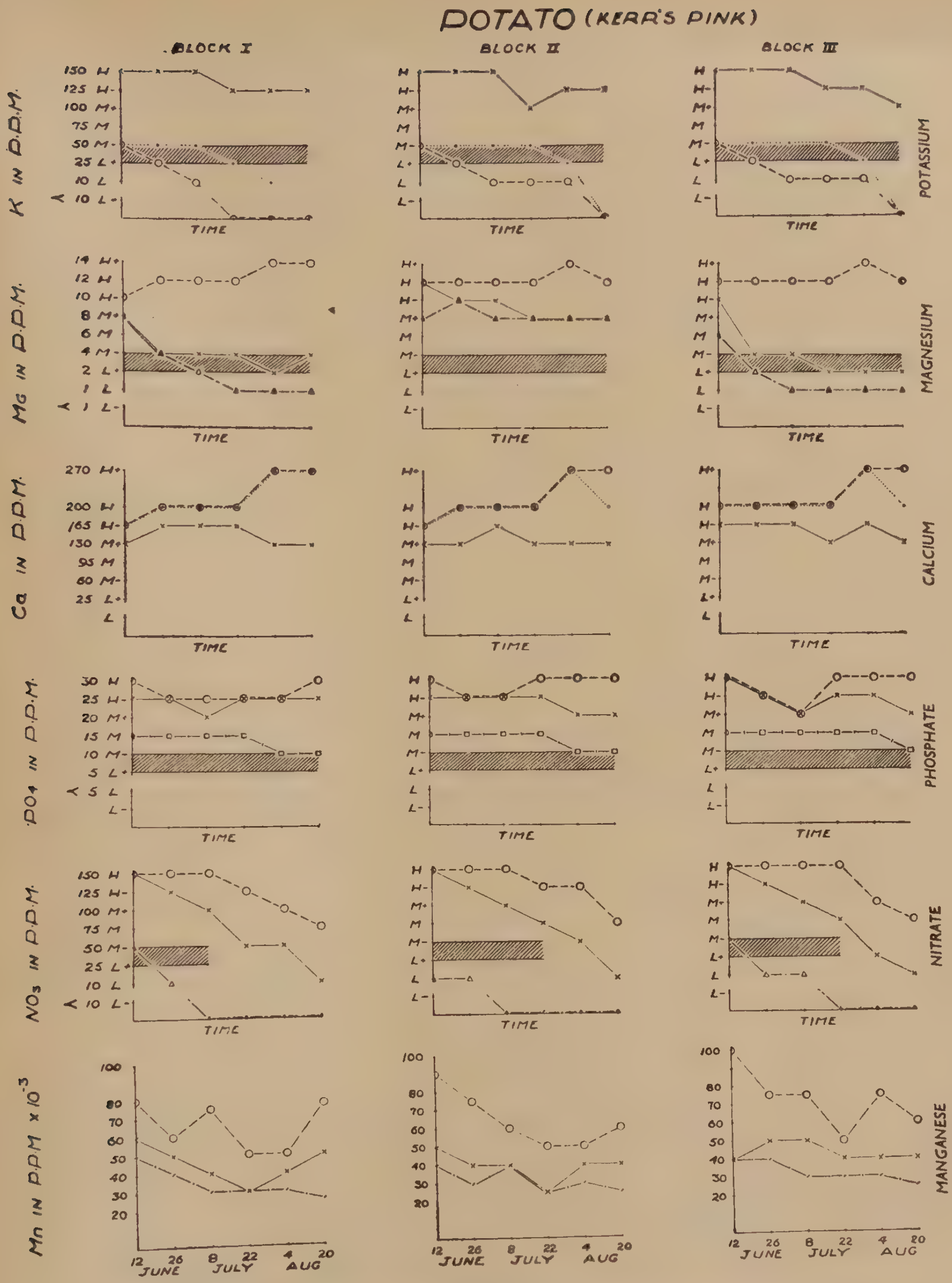

FIG. 8.

The Seasonal Cycles of soluble K, Mg, Ca, PO_4, NO_3 and Mn in POTATO (Kerr's Pink). The following treatments are represented :
(1) ×————× NPK. (2) • . . . . • Nil. (3) ○————○ NP.
(4) △—— – ——△ PK. □——|——□ NK. Season 1946.
The shaded area represents the threshold range associated with visual signs of a deficiency.

SEASON 1945.

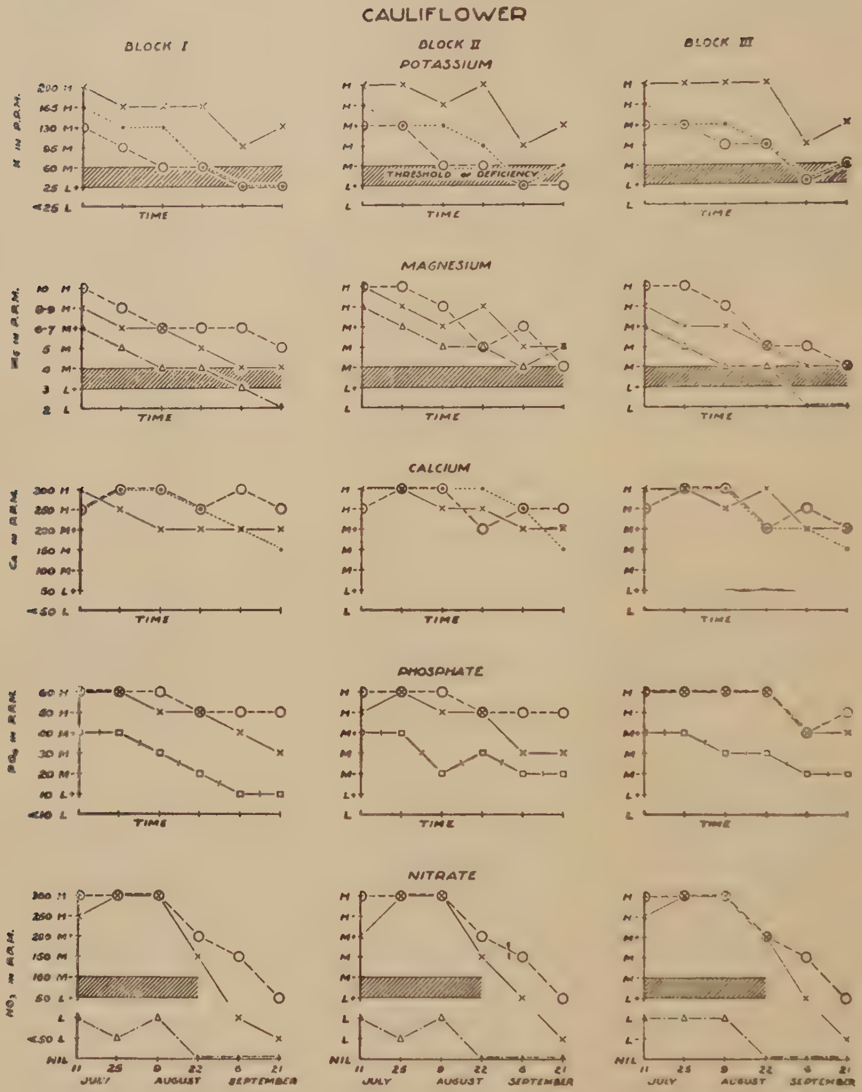

FIG. 9.

The Seasonal Cycles of soluble K, Mg, Ca, PO_4 and NO_3 in CAULIFLOWER. The following treatments are represented:

(1) ×————× NPK. (2) •..........• Nil. (3) ○————○ NP.
(4) △—— - ——△ NP. (5) □——|——□ Season 1945.
The shaded area represents the threshold range associated with visual signs of a deficiency.

Season 1946.

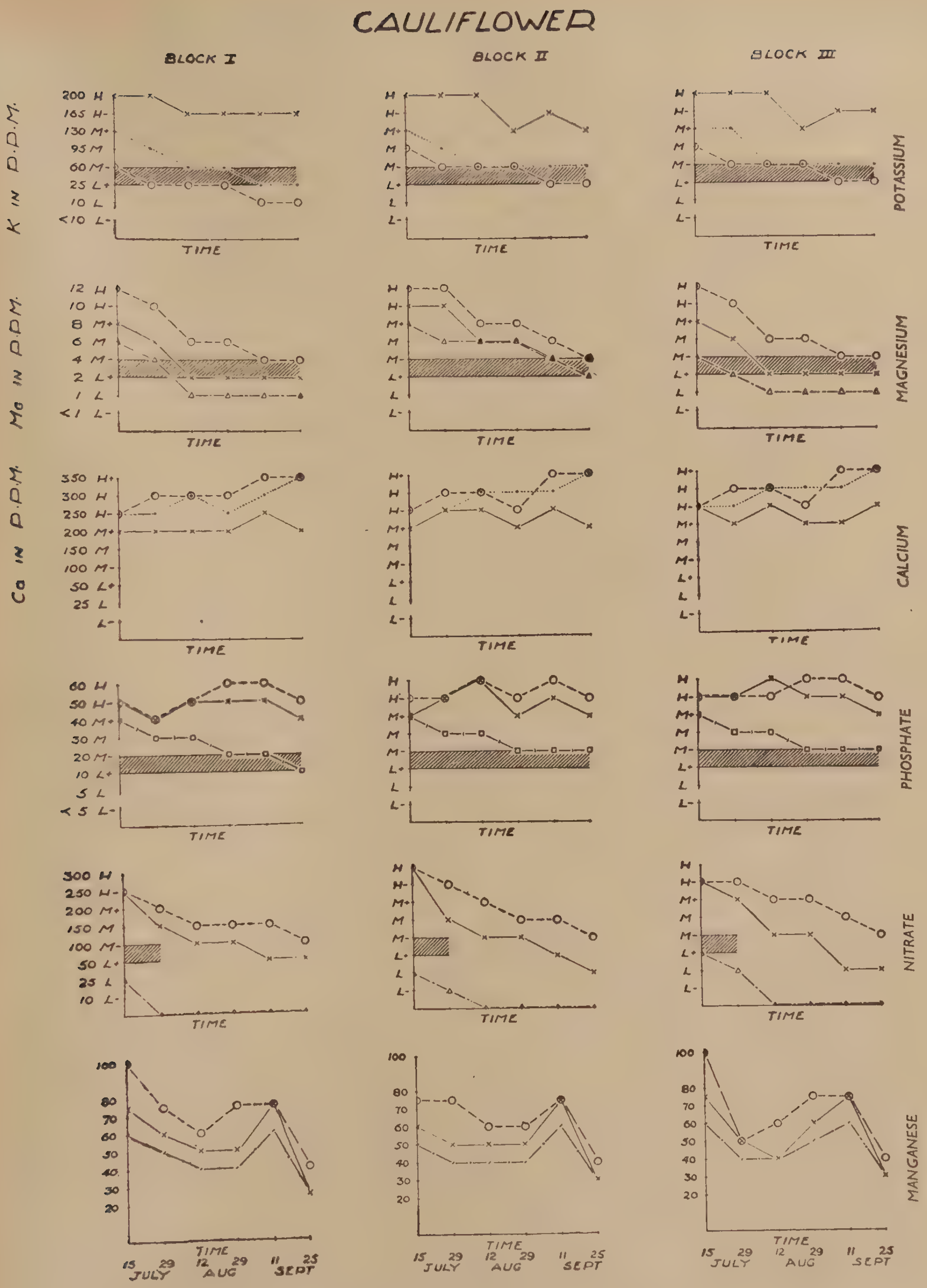

Fig. 10.

The Seasonal Cycles of soluble K, Mg, Ca, PO_4, NO_3 and Mn in Cauliflower.
The following treatments are represented:
(1) ×———× NPK. (2) •..........• Nil. (3) ○———○ NP.
(4) △—— - ——△ PK. (5) □——|——□ Season 1946.
The shaded area represents the threshold range associated with visual signs of a deficiency.

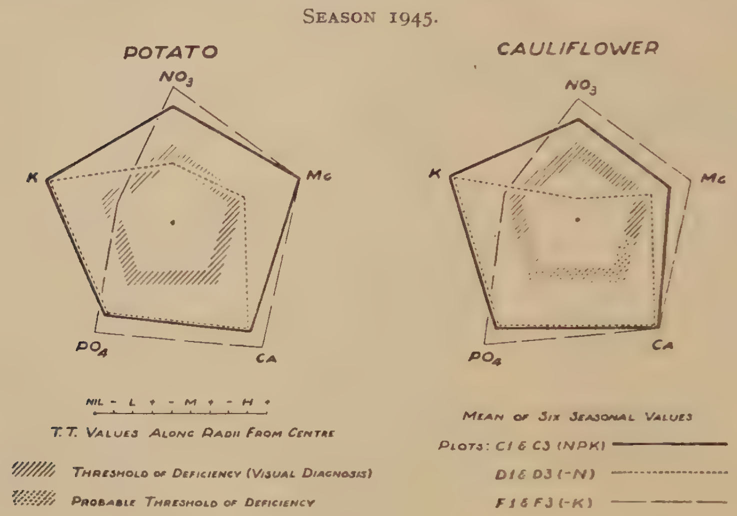

FIG. 11.

Diagrams showing the relation between soluble K, Mg, Ca, PO_4 and NO_3 in POTATO and CAULIFLOWER growing under various manurial treatments (Season 1945). The mean of the six tissue test values for each nutrient for the NPK, NP (Omit K) and PK (Omit N) treatments respectively are plotted along 5 radii of a circle (results for Blocks I and III are bulked together).

which resulted in the leaching out of nitrates from the surface soil. This was confirmed by soil analysis and by the observation that Mg deficiency was particularly severe in waterlogged patches. The Nil and NP (Omit K) plants usually have higher Mg values, thus confirming the reciprocal relation between K and Mg noted in the previous season. The fact that the Mg level in the variety Kerr's Pink is usually lower than in Majestic is in accord with the greater susceptibility of the former to Mg deficiency. The cauliflower usually has a lower Mg level than has the potato.

Calcium. The Ca levels are adequate throughout as might be expected because of the 5 per cent. free lime present in the surface soil. The values are higher when K is omitted, and this is also commonly observed when K is deficient.

Phosphorus. The Omit P treatment is reflected in the NK plants. The absence of K tends to increase the P levels in the tissues.

Nitrate nitrogen. There is a significant difference between treated and untreated N plants during both seasons. The lower values in 1946 may be due to persistent rainfall leaching nitrate N from the top soil. In the absence of K, nitrate values are higher.

Manganese. When K is omitted as in Nil and NP plants the Mn level is usually higher than in other treatments. In F.Y.M. plants, Mn values tend to be lower than in other treatments. This may be due to biological fixation of Mn which may occur in the presence of a higher level of organic matter in a calcareous soil.

TISSUE TESTS IN RELATION TO THE VISUAL SYMPTOMS AND NUTRIENT CYCLES.

In Figs. 6-10 are given the seasonal trends of the soluble nutrients in the two potato varieties and in the cauliflower for seasons 1945 and 1946. The cross hatched bands which appear in these figs. are termed " Threshold of deficiency (tissue test) levels ". These are fixed by the level at which visual symptoms of the respective nutrient deficiencies first appear. In most instances these threshold levels coincide with the Medium minus to Low plus range but quantitative values for these levels vary with nutrient and crop plant (Table III). In some instances, however, especially late in the season, certain nutrients may fall within this range without characteristic visual signs of the deficiency being observed, e.g. nitrate N. Such low values late in the season result from the natural downward trends of the nutrients.

Shaded bands are omitted from graphs when no well-defined visual signs were observed. In a few instances the critical levels have been determined from experimental data from other centres, e.g. deficiency levels of Ca in potato were fixed from plants exhibiting " acidity " symptoms at Sutton Park (p. 107).

Tissue tests may thus be used to confirm visual symptoms but, in addition, when related to the seasonal cycles, may also indicate an impending mineral deficiecny.

Potassium. The deficiency was noted at an early growth stage in potato during both seasons and appeared first in the NP (Omit K) and later in the Nil plants. Table VI gives the relation between tissue test results and the percentage number of K deficient potato plants.

There is close agreement between the onset of visual symptoms of K deficiency and tissue test results. During seasons 1945 and 1946 symptoms were first observed in the NP treatment on June 25th and June 20th respectively in Majestic, and on June 15th, 1946, in Kerr's Pink. On August 13th and 22nd, 1945 and 1946 respectively, when 100 per cent. of the Omit K potato plants showed symptoms, no soluble potash could be detected in the tissues. In comparing data for the two seasons there is general agreement (1) that K deficiency is most severe in the NP treatments, (2) that the deficiency becomes more severe with season, and (3) that values are in accord with the severity of the symptoms noted.

The values in cauliflower did not fall as low as in potato and visual symptoms were less marked. In potato and cauliflower, medium K values in early May and July respectively were unsatisfactory as, within a week, visual signs of the deficiency appeared. Thus in these instances tissue test results may be used to anticipate the deficiency.

Magnesium. Deficiency symptoms were observed in PK (Omit N) potato in Blocks I and III in early June and July respectively in Kerr's Pink and Majestic. Tissue test values are at the threshold of deficiency at those points in the growth cycle. During 1946 the levels in NPK plants fall within the critical range whereas NP (Omit K) plants, as in the previous year, have higher values than any other treatment. Potato plants grown in Block II were free from symptoms and Mg tissue test levels are usually higher.

Data for cauliflower show that the deficiency is greater when N is omitted and K is applied. Visual signs and tissue test results agree that the deficiency is more marked than in potato, even in Mg treated and NP (Omit K) plants, values fall to within the threshold from mid to late season.

Calcium. There is a marked increase in soluble Ca when K is low. The absence of symptoms and relatively high values accord with the 5 per cent. free lime present in the surface soil.

Phosphorus. Leaf symptoms characteristic of P deficiency did not develop but the spindly growth habit has been observed in NK (Omit P) plants. The tissue test and visual data suggest that this nutrient is near the threshold of deficiency.

TABLE VI.

Relation between the incidence of K deficiency and tissue test values in potato (Majestic and Kerr's Pink) at seasonal intervals in 1945 and 1946 (Block I only).

Treatment:		Nil.		NPK.		NP (Omit K).			Nil.		NPK.		NP (Omit K).	
Variety.	Date.	Percentage number of plants showing visual symptoms.	K Tissue Test.	Percentage number of plants showing visual symptoms.	K Tissue Test.	Percentage number of plants showing visual symptoms.	K Tissue Test.	Date.	Percentage number of plants showing visual symptoms.	K Tissue Test.	Percentage number of plants showing visual symptoms.	K Tissue Test.	Percentage number of plants showing visual symptoms.	K Tissue Test.
Majestic ..	18/6/45	Nil	Medium +	Nil	High	Nil	Medium	12/6/46	Nil	Medium +	Nil	High —	Nil	Medium +
Kerr's Pink									10%	Medium —	Nil	High	25%	Medium —
Majestic ..	2/7/45	Nil	Medium	Nil	High —	48%	Low +	8/7/46	30%	Low +	Nil	Medium +	45%	Low +
Kerr's Pink									22%	Medium —	Nil	High	60%	Low
Majestic ..	13/8/45	100%	Nil	Nil	Medium +	100%	Nil	22/8/46	100%	Nil	Nil	Medium +	100%	Low —
Kerr's Pink									100%	Nil	Nil	High —	100%	Nil

In cauliflower, values fell to low levels in the NK (Omit P) treatment during the 1945 season but no symptoms were noticed. During the second season, however, similar values were associated with trace P deficiency. It is probable that the P level is near the threshold of deficiency.

Nitrogen. The nitrate levels, irrespective of treatment, fall as the season progresses in all crop plants studied and at the late stage of growth there is little correlation between nitrate level and visual signs of N deficiency. After certain points in the growth cycle the nitrate test cannot be used as an indicator of the nitrogen status of plants. Thus in Figs. 6-10 the hatched bands are confined to the early season when differences between treated and non-treated N plants are usually significant and values agree with the effects of N deficiency.

Visual symptoms were noted earlier in the 1946 season in the PK treatment, probably due to the wet, cold periods during the season with consequent inhibition of nitrifying bacteria and leaching out of nitrate from the soil. The lower nitrate status of the soil was confirmed by analysis and also by the observation that N deficiency was particularly severe in wet areas.

Manganese. No symptoms were observed in potato and cauliflower and tissue test values made during the 1946 season appear to be adequate. In potato and cauliflower the F.Y.M. and NP plants usually have the lowest and highest values respectively, and those receiving NPK are intermediate. There is an initial decline followed by an increase later in the season and in cauliflower this is followed by a further decrease in level.

INTERRELATIONSHIPS OF EXTRACTABLE NUTRIENTS.

Attention has been drawn to the critical values which can be related to the onset of visual symptoms of a mineral deficiency. Tissue test data also show that there is a close correlation between certain extractable nutrients in both potato and cauliflower, e.g. low N level with low Mg and low K levels with higher levels of Mg, Ca, PO_4, nitrate N, and Mn. These results are plotted graphically in Fig. 11. The means of the six seasonal results for K, Mg, Ca, PO_4 and NO_3 for Blocks I and III are plotted along 5 equidistant radii of a circle for the NPK, PK and NP treated potato and cauliflower. The cross hatched area in each diagram represents the threshold of deficiency as determined by visual diagnosis at centre A; the stippled portion represents the threshold as determined by data from other centres.

Potato (*Majestic*) 1945. The relation between the soluble nutrients K, Mg, Ca, PO_4 and NO_3 in the NPK plants is represented by a symmetrical diagram placed well outside the band showing threshold of deficiency. In the PK (Omit N) plants, however, the mean NO_3 and Mg results are within the threshold, showing the association between low N status and low Mg level. In the NP (Omit K) plants the mean K level is within the deficiency range but Ca, PO_4, and NO_3 are higher than in NPK plants.

Cauliflower. The cauliflower diagram is comparable with that of potato, but it shows that the K values are higher and N values lower than in potato. The Mg value is also lower in the cauliflower.

TISSUE TESTS IN RELATION TO YIELD DATA.

Yield data were obtained for seasons 1945 and 1946 for both potato varieties, and for the cauliflower. in all treatments, and these were statistically treated. The results are given in Tables VII and VIII. The symbols for the manurial treatments are as follows:

A=Nil. B=F.Y.M. C=NPK. D=PK (Omit N).
E=NK (Omit P). F=NP (Omit K).

Potato. Yields for total crop are given in Table VII. The results show that the omission of potash is the main factor depressing yields. This is in accord with the visual symptoms and the low tissue values for plants not receiving potash (Figs. 6, 7 and 8). Visual symptoms of magnesium deficiency were noted in the PK (Omit N) plots and confirmed by tissue test and ash analysis but the soil application of $MgSO_4$ to Block II, although correcting the symptoms and increasing Mg content of the foliage, did not affect the yield. The importance of N in the yield

TABLE VII.

Total yields of potato (Majestic and Kerr's Pink) for seasons 1945 and 1946. Showing treatment, total and mean yields respectively (tons per acre) and order of yields.

(*a*) *Majestic 1945.*

Manurial treatment.	A.	B.	C.	D.	E.	F.	Total.	MEAN.
Block I	3·18	11·25	12·19	8·43	7·88	3·0	45·93	7·66
Block II	6·0	9·20	11·06	9·0	10·31	5·43	51·0	8·5
Block III	5·62	9·38	10·13	10·13	7·68	5·06	48·0	8·0
Total in tons ..	14·8	29·83	33·38	27·56	25·87	13·49	144·93	
Mean in tons ..	4·93	9·94	11·13	9·18	8·63	4·5	48·31	
Order of yields ..	5	2	1	3	4	6		

Yields S.E. = 1·25. S.D. 5% = 3·92. S.D. 1% = 5·58.
At 5% point: B.C.D.E.>F. B.C.D.>A.
At 1% point: C.>AF.
Differences between block yields were not significant.

(*b*) *Majestic 1946.*

Manurial treatment.	A.	B.	C.	D.	E.	F.	Total.	MEAN.
Block I	1·69	12·19	9·0	7·31	6·75	3·0	39·94	6·66
Block II	3·94	7·31	9·56	5·25	5·44	6·56	38·06	6·34
Block III	4·88	6·94	10·69	6·38	4·69	4·68	38·26	6·38
Total in tons ..	10·51	26·44	29·25	18·94	16·88	14·24	116·26	
Mean in tons ..	3·50	8·81	9·75	6·31	5·63	4·75	38·75	
Order of yields ..	6	2	1	3	4	5		

Yields S.E. = 1·06 S.D. 5% = 3·37. S.D. 1% = 4·79.
At 5% point: C.>A.D.E.F. B.>A.F.
At 1% point: C.>A.F. B.>A.
Differences between block yields were not significant.

(*c*) *Kerr's Pink 1946.*

Manurial treatment.	A.	B.	C.	D.	E.	F.	Total.	MEAN.
Block I	5·44	14·25	10·5	10·13	12·0	3·56	55·88	9·31
Block II	6·75	12·56	14·63	10·31	13·88	5·63	63·76	10·63
Block III	6·56	13·69	13·5	11·63	11·25	4·31	60·94	10·16
Total in tons ..	18·75	40·5	38·63	32·07	37·13	13·5	180·58	
Mean in tons ..	6·25	13·5	12·88	10·68	12·38	4·5	60·19	
Order of yields ..	5	1	2	4	3	6		

Yields S.E. = 0·56. S.D. 5% = 1·78. S.D. 1% = 2·53.
At 5% point: B.C.>A.D.F. E.>A.F. D.>A.F.
At 1% point: B.>D.A.F. C.D.E.>A.F.
Differences between block yields were not significant.

of potato is shown in season 1946 in both Majestic and Kerr's Pink, the yields for the NPK treatments being significantly greater than those of PK (Omit N) treatments.

Cauliflower. The total weights of heads cut for each plot were recorded. The statistical analysis is given in Table VIII.

The results emphasize that N deficiency is the main factor in depressing head formation. Tissue test results and visual symptoms both confirm that N deficiency was severe after the first sampling dates of the two seasons, viz. July 11th, 1945, and July 15th, 1946 (Figs. 9 and 10), when nitrate N in PK (Omit N) plants was within the deficiency limit. The addition of $MgSO_4$ to Block II, although alleviating Mg deficiency, as shown by visual and chemical data, had no significant effect on the yields. Potash deficiency had no depressing effect on head formation which is in accord with the milder visual effects and higher tissue test levels recorded during the season in cauliflower as compared with potato under Omit K treatments.

TISSUE TESTS IN RELATION TO ASH ANALYSIS OF THE FOLIAGE.

The correlations between tissue test and ash analysis data for K, Mg, Ca, P and Mn are given for potato (Majestic) and cauliflower for seasons 1945 and 1946 in Figs. 12-15. The tissue test data on the ordinates in parts per million are plotted, irrespective of treatment, against ash analysis on the abscissae as percentage in dry matter. A total of 54 comparisons is possible for the three seasonal sampling times. The figures above the points indicate the number of times the chemical values coincide. The main points are as follows:

Potassium. The two methods are in agreement over the range studied. The data suggest that 1 to 2·5 per cent. K_2O in dry matter approximates to the tissue test threshold of deficiency range, viz. 25 to 50 p.p.m. K. These values are associated with the development of visual signs of the deficiency in potato and cauliflower.

Magnesium. The series of points fall on a line indicating a close correlation. In potato the critical L+ and M− tissue test range corresponds with 0·25 to 0·5 per cent. MgO, and at these levels visual signs of Mg deficiency appear. The fact that the high tissue test value (11 p.p.m.) is applicable to 1 to 2 per cent. MgO shows that the tissue test method is not sensitive at high levels.

TABLE VIII.

Total yields of cauliflower for the seasons 1945 and 1946. Showing treatment, total and mean yields respectively (tons per acre) and order of yields.

(*a*) *1945 Season.*

Manurial treatment.	A.	B.	C.	D.	E.	F.	Total.	MEAN.
Block I ..	2·62	3·55	4·75	2·16	3·80	3·95	20·83	3·47
Block II ..	4·17	4·07	4·23	2·44	3·49	3·91	22·31	3·72
Block III ..	2·93	4·38	4·04	2·81	3·36	4·41	21·93	3·66
Total in tons ..	9·72	12·00	13·02	7·41	10·65	12·27	65·07	
Mean in tons ..	3·24	4·0	4·34	2·47	3·55	4·09	21·69	
Order of yields ..	5	3	1	6	4	2		

Yields S.E. =0·36. S.D. 5% =0·8. S.D. 1% =1·14.
At 5% point: B.C.E.F.>D.
At 1% point: B.C.F.>D.
Differences between block yields were not significant.

(*b*) *1946 Season.*

Manurial treatment.	A.	B.	C.	D.	E.	F.	Total.	MEAN.
Block I ..	3·52	7·09	5·63	2·11	3·97	5·60	27·92	4·65
Block II ..	3·20	5·42	4·94	3·44	5·59	7·41	30·00	5·00
Block III ..	2·19	6·0	6·04	3·53	6·20	8·06	32·02	5·34
Total in tons ..	8·91	18·51	16·61	9·08	15·76	21·07	89·94	
Mean in tons ..	2·97	6·17	5·54	3·03	5·25	7·02	29·98	
Order of yields ..	6	2	3	5	4	1		

Yields: S.E. =0·52. S.D. 5% =1·36. S.D. 1% =1·93.
At 5% point: F.B.C.E.>AD. F.>C.E. At 1% point: F.B.C.E.>A.D.
Differences between block yields were not significant.

In cauliflower the tissue test deficiency threshold L+ to M− (2 to 4 p.p.m.) may be referred to approximately 0·25 per cent. MgO in dry matter.

Calcium. The two methods agree in showing that the Ca status is adequate throughout and that the M+ to H+ (130 to 200 p.p.m.) range corresponds to 4 to 8 per cent. CaO in potato whereas cauliflower has higher tissue test values for a similar total range.

Phosphorus. Tissue test data for PO_4 in parts per million, as ordinates, are referred to total values expressed as percentage P_2O_5, as abscissae.

The results for potato suggest that the two methods are related over the whole range and that 0·25 per cent. P_2O_5 corresponds to the L+ to M− (10 to 15 p.p.m. PO_4) which may be characterized by the onset of symptoms. In cauliflower approximately 0·5 per cent. P_2O_5 corresponds to the L+ to M− tissue test range but no definite visual signs were noted.

Manganese. The soluble Mn levels in parts per thousand million (p.p.m. × 10^{-3}) on the ordinate are referred to the total Mn content in the ash in parts per million dry matter on the abscissae. The quantitative data are more exact than for major elements and thus a greater scatter of points is permissible for each category.

In potato and cauliflower the results of the two methods conform to a line. A given value for soluble Mn in p.p.m. × 10^{-3} may within experimental error be referred to a similar total value in parts per million. The range covered is from approximately 20 to 70 × 10^{-3} p.p.m., soluble, and 20 to 70 p.p.m., total. Data from other centres suggest that 10 to 20 × 10^{-3} p.p.m., soluble, and 10 to 20 p.p.m., total, approximate to the deficiency level characterized by development of visual symptoms.

SEASON 1945.

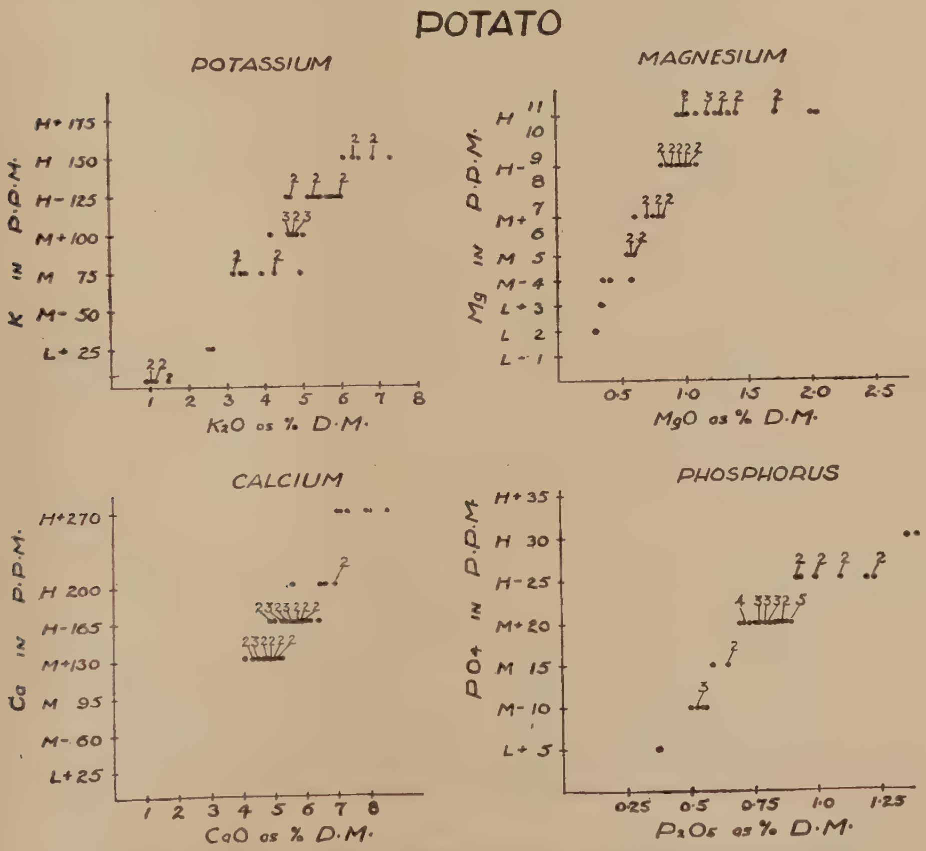

FIG. 12.

The Relation between Tissue Tests (ordinates) and Ash Analyses (abscissae) for K, Mg, Ca and P in POTATO (Majestic). Tissue test and ash analysis data given in parts per million (p.p.m.) in the extract and as percentage dry matter (D.M.) respectively. Data for the three seasonal sampling times are plotted irrespective of treatment. Season 1945. Figures above points indicate the number of times values coincide.

Season 1946.

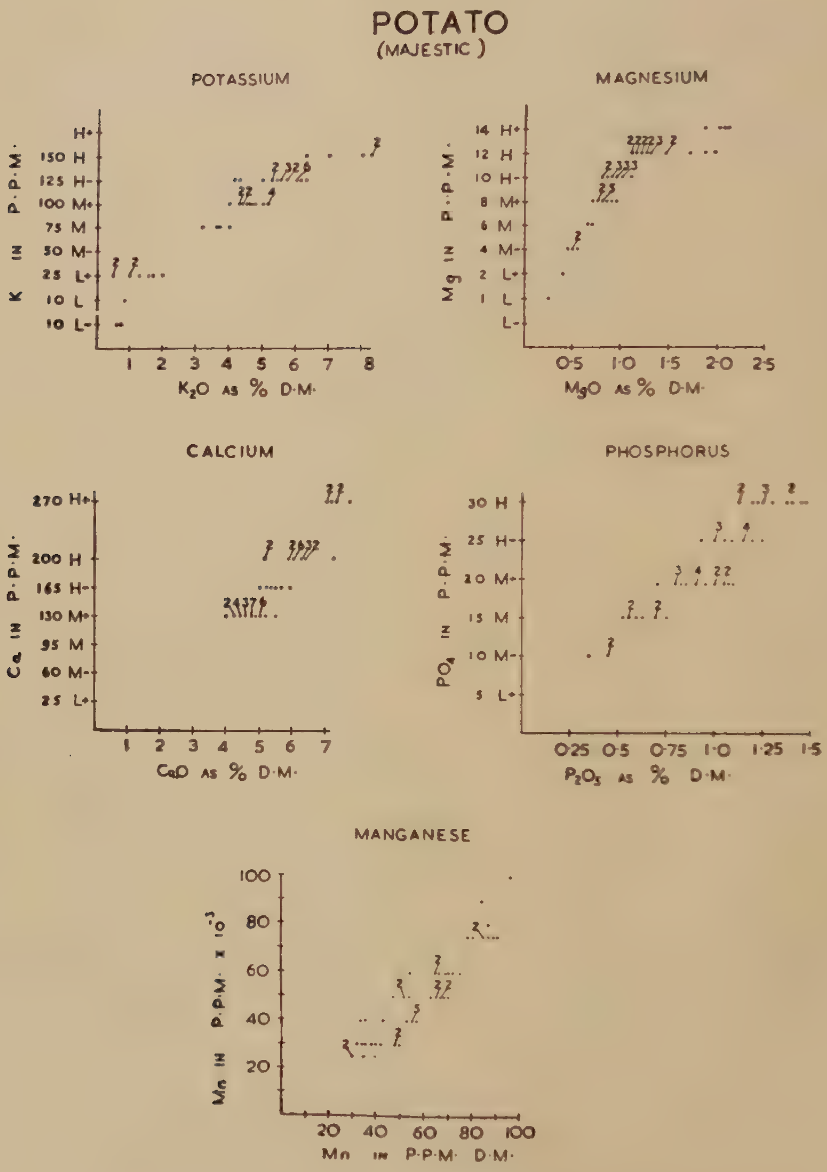

Fig. 13.

The Relation between Tissue Tests (ordinates) and Ash Analyses (abscissae) for K, Mg, Ca, P and Mn in Potato (Majestic). Tissue test and ash analysis data given in parts per million (p.p.m.) in the extract and as percentage dry matter (D.M.) respectively. Mn in p.p.m. in the extract and dry matter. Data for the three seasonal sampling times are plotted irrespective of treatment. Season 1946. Figures above points indicate the number of times values coincide.

SEASON 1945.

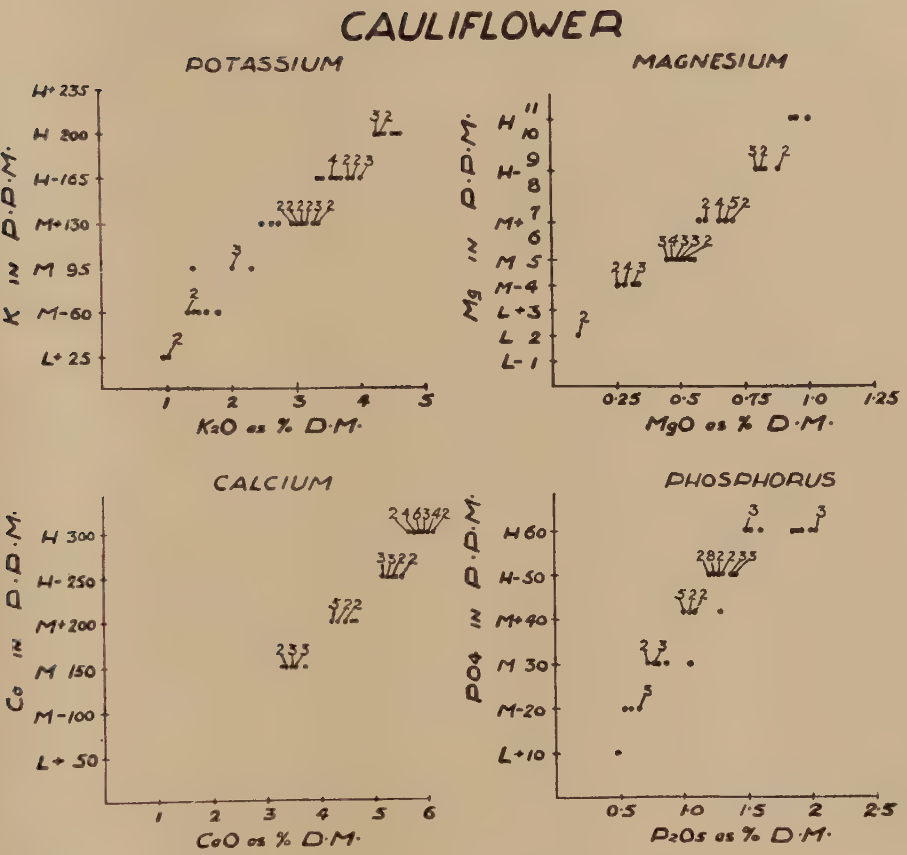

FIG. 14.

The Relation between Tissue Tests (ordinates) and Ash Analyses (abscissae) for K, Mg, Ca, and P in CAULIFLOWER. Tissue test and ash analysis data given in parts per million (p.p.m.) in the extract and as percentage dry matter (D.M.) respectively. Data for the three seasonal sampling times are plotted irrespective of treatment. Season 1945. Figures above points indicate the number of times values coincide.

SEASON 1946.

CAULIFLOWER

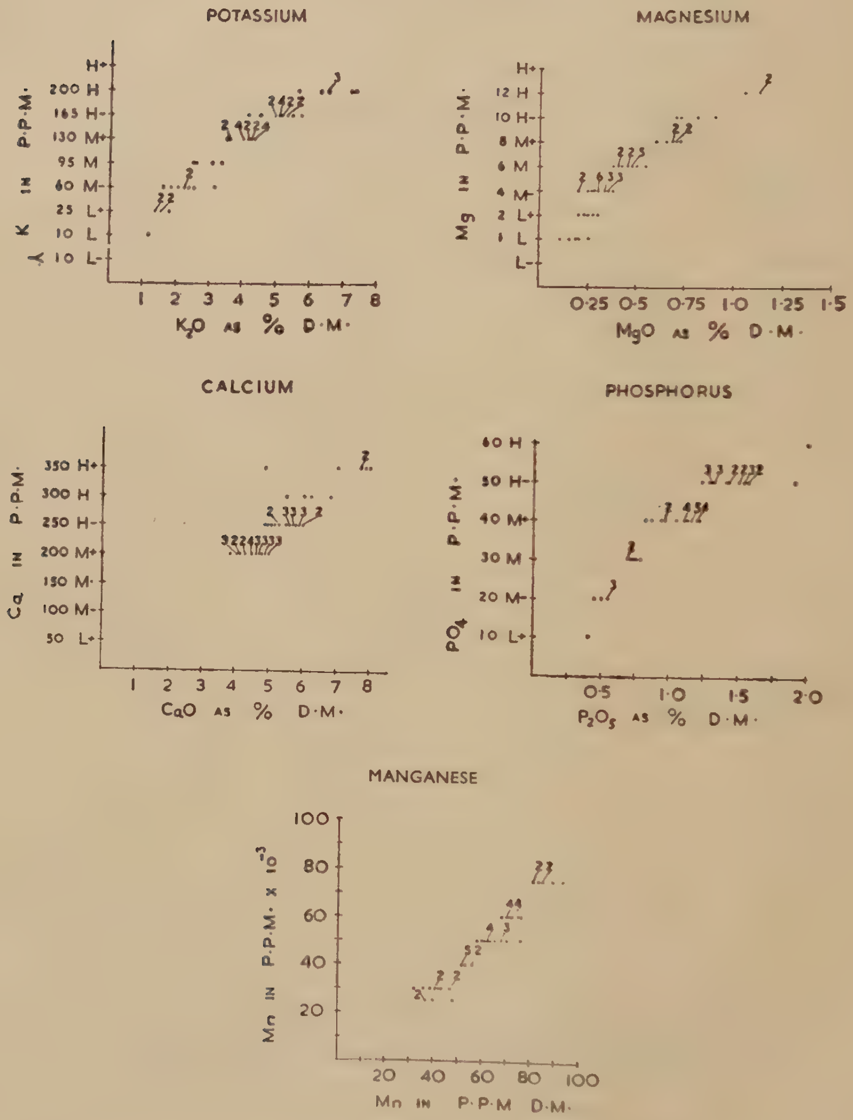

FIG. 15.

The Relation between Tissue Tests (ordinates) and Ash Analyses (abscissae) for K, Mg, Ca, P and Mn in CAULIFLOWER. Tissue test and ash analysis data given in parts per million (p.p.m.) in the extract and as percentage dry matter (D.M.) respectively. Mn in p.p.m. in the extract and dry matter. Data for the three seasonal sampling times are plotted irrespective of treatment. Season 1946. Figures above points indicate the number of times values coincide.

DISCUSSION.

A rapid tissue test technique has been developed and used in the quantitative study of the mineral status of K, Mg, Ca, Mn, PO_4 and nitrate N in potato (Majestic and Kerr's Pink) and cauliflower grown on a long term manurial trial.

In using the method a number of precautions must be taken. It is essential to standardize the field sampling procedure and to use special parts of plants that are morphologically similar. Standards for the soluble nutrients at High, Medium, and Low levels were fixed for the plants examined and the quantitative values for each level were found to vary from one crop plant to the other. It was necessary to carry out a number of preliminary determinations on healthy plants and on others showing visual signs of a mineral deficiency or toxicity to fix the quantitative values for each category. Tissue test values must be referred to the seasonal cycles for the nutrients as is shown in the data for samples taken at fortnightly intervals during the growing season. By the use of such data a deficiency or a toxicity effect can be distinguished from a seasonal trend characteristic of the crop plant and nutrient. Thus nitrate values fall to lower levels with the progress of the season in most crop plants, irrespective of treatment, and tissue test values cannot be used after certain points in the growth cycle for the diagnosis of N deficiency. The decline of nitrate N with season has also been reported by Chibnall (5), Vaidya (48) and Nightingale (36, 37) working with grass, apple and pineapple respectively.

A study of the seasonal cycles of the soluble nutrients is important for diagnostic work, e.g. medium values for certain mineral elements such as K and Mg, during early season, may suggest an impending deficiency in a susceptible crop, viz. potato and cauliflower, whereas similar results late in the season may be normal values for healthy crops, resulting from the natural downward seasonal trends characteristic of crop and nutrient. Tissue test data may be used to anticipate mineral deficiencies or toxicities prior to the development of visual symptoms.

Tissue test data show that on experimental plots the effects of the different fertilizer treatments may be recognized in the crops by this method and also that different reactions of crops growing on the same plot may be demonstrated, e.g. the K and Mg levels in potato were usually lower and higher respectively than for cauliflower grown under the same manurial treatments.

Correlation studies between tissue test results and those of the visual diagnosis method (49) were made and in these it was found possible to relate tissue test values for K, Mg, P, nitrate N and Mn, to the onset of visual symptoms. These values are regarded as threshold values for the particular deficiencies or toxicities.

Yield data for 1945 and 1946 show that K and N deficiencies are mainly responsible for the depression in yields of potato and cauliflower respectively. These results agreed with the low tissue values for K and nitrate N obtained during the growing period.

Although Mg deficiency was indicated in the PK (Omit N) treatments by visual symptoms, tissue tests and ash analysis, there were no significant differences in yields as compared with those receiving Mg, where symptoms were absent, and chemical values for Mg were higher. This demonstrates that yields do not always reflect a mineral deficiency effect.

The great value of tissue tests would be to replace with quick chemical methods the tedious and time-consuming procedures used in "total" analysis and thus several comparisons of the two procedures have been made. At Long Ashton, tissue test data for potato and cauliflower were related to total analysis at three points in the 1945 and 1946 seasons respectively. The correlation diagrams show that there is a linear relation between the results of the two methods over the critical diagnostic range for deficiencies. At high levels found in healthy plants, however, the tissue test method does not usually distinguish different levels, due either to the fact that the 15 minutes period for the extraction of soluble nutrients is not long enough to distinguish between these high values or that over a certain total content, the proportion of less easily soluble material increases rapidly, or that at such high levels it is more difficult to differentiate colour and turbidity differences than at a lower range. For diagnostic work to determine the causes of unsatisfactory crops the point is not important and for the ranges used in both mineral deficiency and toxicity problems agreement between the two methods is close. The quick tissue tests may therefore replace the longer " total " determinations for the purpose discussed in this paper. Finally it may be stressed that the tissue test method is not only rapid and simple in operation but can be used in the field and hence is a valuable adjunct to visual and other field methods for advisory and survey work.

SUMMARY.

1. A rapid chemical tissue test method is described which has been used for the diagnosis of K, Mg, P, N and Mn deficiencies in potato (two varieties) and cauliflower, grown on a long term manurial trial.

2. The tissue test data reflected the effects of manurial treatment and also showed differences in the nutrient status of different crops grown on the same plot, e.g. potato showed a lower K content than cauliflower on plots receiving NP (Omit K) manuring.

3. The results obtained by the method showed good agreement with visual diagnosis and it was possible to fix threshold values at which deficiencies of K, Mg, P and N produced visual symptoms in a number of crops.

4. To use tissue test values for diagnostic purposes it is necessary to relate them to the seasonal cycles of the various nutrients for particular crops. Thus nitrate N was found to fall continuously as the season progressed and the nitrate test could not be used to determine N status in potatoes and cauliflower late in the season. On the other hand, with a knowledge of these cycles, the method could be used to anticipate the development of deficiencies and toxicities which only became visible at a later stage of growth.

6. Field data for potato obtained at one centre showed that K and N deficiencies caused significant reduction in crop weight. In cauliflower, N deficiency was mainly responsible for low yields. These deficiencies were indicated early in the season by chemical tissue tests.

7. A close correlation was found between the data of tissue tests and those of full chemical analysis over the range of values useful for the diagnosis of deficiencies. At the higher nutrient levels found in healthy plants the tissue test method as used did not always distinguish the differences shown by full analysis. The comparisons suggest that the tissue test method may be used to replace the longer procedure for rapid diagnostic purposes.

8. The tissue test method, because of its speed and ease of application and its suitability for use in the field, is particularly valuable for advisory and survey work.

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(*Received* 23/2/48.)

THE APPLICATION OF RAPID CHEMICAL TESTS TO THE DIAGNOSIS OF MINERAL DEFICIENCIES IN HORTICULTURAL CROPS

II. CROPS GROWN AT VARIOUS CENTRES

By **D. J. D. NICHOLAS**
Long Ashton Research Station, Bristol

SYNOPSIS

Further evidence is given of the value of quick chemical tests in the diagnosis of potassium, magnesium, calcium, phosphorus and nitrogen deficiencies and manganese and chloride toxicities in a variety of crops, viz. potato, tomato, black and red currant and apple, grown at widely different sites. The tissue test results showed good agreement with those of other diagnostic methods and were of value in resolving the following complex nutrient disorders: (1) acidity complex in potato where potassium, magnesium, calcium, phosphorus and nitrogen deficiencies and manganese toxicity occurred together; (2) potassium deficiency and chloride toxicity in black and red currant; (3) potassium and magnesium deficiencies in apple.

In the previous paper it has been shown that using the tissue test method the mineral status of potato and cauliflower could be determined at various points in the growth cycle. It has also been demonstrated that the correlation between the results of the tissue test method and those of other diagnostic procedures is close.

An account is given in this paper of the application of tissue tests to the study of the mineral status of a number of horticultural crops including potato, cauliflower, black currant, red currant and apple grown under widely different conditions. It is shown that the correlation between the tissue test data and those of the visual method (6) spray treatment (3) and ash analysis of the foliage is usually good. The rapid tissue test method may be used to diagnose nutritional disorders in a variety of crop plants grown on widely different soil types.

EXPERIMENTAL.

Materials.

The crops were grown at a number of centres under a variety of manurial treatments. Data shown in Table I concern sites, soil characters, crops grown, and the nutritional problems occurring at each centre, as well as the sampling times for tissue tests and ash analysis.

The following supplementary notes are appended for each centre:

Centre A.

The experimental area is a reclaimed acid heathland, originally with a soil pH of approximately 4·0. It was divided into strips to which different rates of ground limestone had been applied in 1944 and across which various fertilizer treatments consisting of NPK combinations are given annually. The rate of application of ground limestone is at 3 and 9 tons per acre; of nitrogen (N), as sulphate of ammonia, at 3 cwt. per acre; of phosphorus (P), as triple superphosphate, at 2½ cwt. per acre and of potash (K), as muriate of potash, at 2½ cwt. per acre.

TABLE I.

Centre.	Sampling dates for tissue tests and ash analysis.	Soil characters.	Crop and variety.	Nutritional problems.
A. Sutton Park, Birmingham.	30.7.46	Highly podsolized gravelly sand, mainly derived from glacial deposits of Bunter origin.	Potato, Majestic.	N, P, K, Mg and Ca deficiencies and Mn toxicity.
B. Glasshouse Experiments in Somerset.	14.11.44 5.12.44	Glasshouse soil, high in N, P and K but low in Mg, derived from calcareous inferior oolite.	Tomato, Ailsa Craig.	Mg deficiency.
C1. Long Ashton Fruit Plots.	17.8.44	As for A.	Black currant, Seabrooks. Red currant, Laxton's No. 1.	K deficiency ; Cl toxicity.
C2 ,,	27.8.44 26.8.45	,,	Apple, Lane's Prince Albert.	K, Mg and P deficiencies.
C3 ,,	7.9.44 9.9.45	,, but has calcareous parent material mainly Keuper Marl.	Apple, Allington Pippin and Edward VII.	K and Mg deficiencies.
D. Kent	4.9.44 26.9.45	Calcareous, sandy loam derived from glacial sludge over chalk.	Apple, Cox's Orange Pippin.	Mg deficiency.
E. Sussex	6.9.44 1.9.45	Light sandy loam, over Lower Greensand.	Apple, Cox's Orange Pippin.	Mg deficiency.
F. Cheshire.	12.9.44	Podsolized sand from mixed red drift.	Apple, Cox's Orange Pippin.	Mg deficiency.
G. Surrey	7.9.44	Light sandy soil derived from Lower Greensand.	Apple, Miller's Seedling.	Mg deficiency.

Centre B.

At this centre, experiments were designed to test the efficiency of control measures to overcome magnesium deficiency in glasshouse tomatoes. The following treatments were given in addition to the grower's NPK base dressing :

1. $MgSO_4$ at 5 cwt. per acre to the soil.
2. $MgSO_4$ at 10 cwt. per acre to the soil.
3. $MgSO_4$ at 15 cwt. per acre to the soil.
4. $MgSO_4$ at 20 cwt. per acre to the soil.
5. $MgSO_4$ as a 2 per cent. foliage spray applied five times, at fortnightly intervals, during the season.
6. $MgSO_4$ as a 2 per cent. solution applied to the soil around the roots ; rate and time of applications as for foliage spray.
7. Nil.

Soil analysis showed a high available K and P and an adequate lime content, but the Mg level was low.

Centre C.

Experiment 1. This trial was planned to test the effects of various types of potash manures on black currants and other fruit crops. The treatments represented are N,P, KCl; N,P,K_2SO_4; and NP (Omit K). The fertilizers were applied annually, nitrogen as nitro-chalk at 2 cwt. per acre, phosphorus as superphosphate (18 per cent.) at 3 cwt. per acre, and potassium as muriate or sulphate of potash at 3 cwt. per acre.

Experiment 2. This comprises a well-established manurial trial on apple, variety Lane's Prince Albert. The trial is laid down in two blocks with the following randomized treatments within each block: nil; farmyard manure (F.Y.M.); complete fertilizer (NPK); PK (Omit N); NK (Omit P); NP (Omit K). The rates of fertilizer application have remained unchanged since 1923, nitrogen as nitrate of soda being applied at 2 cwt. per acre, phosphorus as superphosphate at 3 cwt. per acre, and potassium as muriate of potash at 1 cwt. per acre.

In addition, soil applications and foliage sprays of magnesium sulphate have been given on certain plots to counteract the effects of Mg deficiency. Details of treatments are given, with the results, in Tables V and VI.

Experiment 3. This manurial trial, started in 1921 and using Allington Pippin and Edward VII, was designed to compare the effects of similar NPK dressings applied to different plots in spring, summer and autumn respectively. From 1936 to 1938 half the area, including a complete range of treatments, was given no manure and, from 1939, N only (Omit P and K).

Centres D, E, F and G.

These centres are widely separated and site, climate, soil type and manurial and cultural treatments vary considerably. At each, magnesium deficiency has been serious, and experiments to test control measures have been in progress over a number of seasons. Magnesium sulphate has been used for soil application and stem injection and foliage spray treatments were given at all centres; magnesian limestone was also tested as a soil dressing at centre G.

METHODS FOR CHEMICAL TISSUE TESTS AND ASH ANALYSIS.

Selection of plant parts.

Details of the sampling procedure used for potato, tomato, cauliflower, apple and black currant have been given in the previous paper (p. 74).

The preparation of test samples, extraction of soluble nutrients and the chemical tests used, have also been given in the previous paper (pp. 74-78).

RESULTS.

Nutrient standards. These have been reported in the previous paper (pp. 77, 78, Tables III and IV).

Centre A.

This centre is a reclaimed acid heathland and both liming and NPK manuring are necessary before a potato crop can be grown successfully. The potato plants that survived in unlimed plots generally showed acute " acidity " symptoms and remained stunted. It has been shown recently that Mn tends to accumulate in potato foliage growing under acid conditions and that toxicity levels may be reached (1, 2). Calcium deficiency and manganese toxicity are two of the main factors responsible for the typical " acidity " symptoms in potato (7).

The chemical data, vigour marking and symptoms noted are included in Table II. Vigour markings (maximum=10), based on the level of a good crop, and a record of visual symptoms were made. Low markings are associated with low calcium and high manganese levels in the plots not limed and with low levels of magnesium, potassium and phosphorus in the limed group. The main points are as follows :

Potassium. In the group not limed, values, irrespective of treatment, vary from M— to M (soluble) ; 1·8-3·0 per cent. K_2O (total) and these levels are near the threshold of deficiency. The habit recorded, i.e. squat plants with short internodes, may be partly due to low Ca and high Mn levels in the tissues. There is a greater differentiation of the K treatments in the limed group when treated K plants have a H or H— tissue level and approximately 5·0 per cent. K_2O, and deficient plants have L+, 25 p.p.m. (soluble) and 1·23 per cent. K_2O (total). In most instances the two chemical methods confirm K deficiency. The main discrepancies between the two chemical methods are in plots 8 and 16 where tissue values are above the threshold level whereas full chemical data are below. The threshold of deficiency levels suggested for potato (centre A), tissue test L+ to M— (25-50 p.p.m. K) and ash analysis 1·0-2·5 per cent. K_2O, are confirmed.

Magnesium. The deficiency occurs when K is applied or N omitted as in plots 2-5 and 10-13 with tissue values at L+ to M— and total results between 0·30 and 0·4 per cent. MgO. Values increase when K is omitted ; thus K treatment, under these conditions, accentuates Mg deficiency. Lime (plot 9) also enhances the deficiency as compared with no lime (plot 1) and chemical data are in agreement with symptoms produced. The threshold of deficiency levels at L+ to M— and 0·25 to 0·35 per cent. MgO are comparable to those at other centres.

Phosphorus. In the group not limed, the NPK treatment has a high tissue test value and the full chemical result is in agreement. In plots 1, 5 and 8 the M— values are near the threshold of deficiency and visual signs were not observed, probably due to the masking effect of the other deficiency symptoms which are also present. Chemical results suggest that liming has depressed the phosphate content. The deficiency was observed in plots 12 and 13, in the NK treatments, where values at L+, and 0·28 per cent. P_2O_5 confirm the visual signs.

Calcium and Manganese. In the non-limed group the tissue levels are at L+ and total at 0·63-0·82 per cent. CaO, thus confirming calcium deficiency symptoms observed. In the limed strips the calcium levels vary from M to H— and total at 4 per cent. CaO. The highest values are recorded when K is deficient.

The pH of the soil in the unlimed strips is approximately 4·0, denoting strongly acid conditions. Under such conditions it has been previously reported that manganese can accumulate to such an extent as to produce toxic effects.

The soluble Mn values in the unlimed series are very high and the totals at 300-600 p.p.m. confirm that they are producing toxic effects. The typical small

TABLE II.

Relation between vigour marking, visual symptoms, and full chemical and tissue test data for K, Mg, Ca, P, Mn.

Plot.	Manurial treatment.	Crop vigour max. = 10.	Visual symptoms.	Full chemical data.					Tissue test data.				
				As % dry matter.				In parts per million Mn.					
				K_2O.	MgO.	CaO.	P_2O_5.		K	Mg.	Ca.	PO_4.	Mn.
1	Nil (no lime)	1	Ca deficiency	3·06	0·64	0·70	0·32	390	M	H−	L+	M−	H++
2	NPK ,,	2	Ca and Mg deficiency and Mn toxicity	2·01	0·30	0·82	1·12	540	M−	L+	L+	H	H++++
3	PK ,,	2	Ca and Mg deficiency and Mn toxicity	2·18	0·42	0·54	0·57	457	M−	M−	L+	M	H++
4	NK ,,	3	Ca deficiency and Mn toxicity	2·56	0·37	0·78	0·62	303	M−	M−	L+	M	H++
5	K ,,	2	Ca and Mg deficiency and Mn toxicity	3·82	0·43	0·63	0·38	326	M	M−	L+	M−	H++
6	NP ,,	3	K deficiency habit Mn toxicity	2·89	0·62	0·96	0·53	485	M	M+	M−	M	H++++
7	P ,,	3	K deficiency and Mn toxicity	2·76	0·65	1·0	0·57	620	M−	H−	M−	M	H++++
8	Nil ,,	1	K deficiency habit Mn toxicity	1·88	0·50	0·80	0·40	639	M	H−	L+	M−	H++++
9	Nil+Lime 9 ton per acre	2	Mg deficiency	4·12	0·36	4·01	0·58	130	H−	M	M	M	Tr.++
10	NPK ,, ,,	6	Mg deficiency	5·62	0·40	4·52	0·63	141	H−	M−	M+	M+	Tr.++
11	PK ,, ,,	5	Mg deficiency	4·85	0·30	3·62	0·42	128	H−	L+	M	M	Tr.++
12	NK ,, ,,	4	Mg and P deficiency	5·24	0·42	3·92	0·28	117	H−	M−	M+	L+	Tr.++
13	K ,, ,,	3	Mg and P deficiency	5·86	0·52	4·06	0·30	136	H	M	M+	L+	Tr.++
14	NP ,, ,,	4	K deficiency habit	1·23	0·63	5·02	0·46	152	L+	M+	H−	M−	Tr.+
15	P ,, ,,	2	K deficiency	1·78	0·58	5·36	0·30	156	L+	M+	H−	L+	Tr.+
16	Nil ,, ,,	1	K deficiency habit	2·12	0·48	5·42	0·28	162	M	M	H−	L+	Tr.+

leaf effect and stunted growth associated with Ca deficiency (6) were recorded, as well as Mn toxicity symptoms, i.e. necrotic brown patches and tinting around the main vein, with occasional black lesions on the petiole and stem. The latter symptom was particularly marked when Ca and K were omitted as in 6, 7 and 8. Chemical results suggest that this symptom is associated with low calcium and high manganese levels with K and Mg also near the threshold of deficiency levels. When lime is applied, the soluble and total Mn levels in the plants are reduced to Tr.++ (0·8 p.p.m.) and 117-162 p.p.m. Mn respectively. Thus the lime treatment not only supplies the required Ca but also controls the intake of manganese by the plants.

Centre B.

Details of the incidence of Mg deficiency in the tomato crops under various Mg treatments are given in Table III. In addition, tissue test and ash analysis values are included for the two sampling times during the growing season.

TABLE III.

Percentage Mg deficient plants and the mean tissue test and ash analysis data for K, Mg, Ca and P for duplicate treatments on tomatoes.

Date of sampling.	$MgSO_4$ treatments.	Mean % Mg deficient plants in duplicate treatments.	Tissue test data.				Ash analysis as % dry matter.			
			K.	Mg.	Ca.	PO_4.	K_2O.	MgO.	CaO.	P_2O_5.
14.11.44	5 cwt. per acre to soil.	23	H	L+	H	H	4·67	0·39	10·52	0·95
	10 cwt. ,,	7	H−	M−	H	H	4·12	0 51	10·00	0·80
	15 cwt. ,,	9	H	M−	H	H	4·83	0·52	11·38	0·81
	20 cwt. ,,	7	H	M−	H	H	4·68	0·43	11·62	0·84
	2% foliage spray applied 5 times during season.	Nil	M+	H+*	H	H	3·78	1·75*	9·42	0·95
	2% liquid applied to soil around roots.	19	H	L+	H	H	4·95	0·39	11·00	0·97
	Nil.	38	H	L−	H	H−	4·92	0·10	9·50	0·75
5.12.44	5 cwt. per acre to soil.	26	H	L+	H	H−	5·10	0·38	11·32	0·79
	10 cwt. ,,	13	H	L+	H	M+	5·26	0·41	9·76	0·75
	15 cwt. ,,	16	H	M−	H	H−	4·63	0·52	11·78	0·80
	20 cwt. ,,	15	H	M−	H+	H−	4·65	0·42	10·85	0·79
	2% foliage spray	Nil	M+	H+*	H	H−	3·76	1·90*	10·78	0·75
	2% liquid to soil	25	H	L+	H	H	5·57	0·26	10·23	0·89
	Nil	43	H	L−	H	H−	5·81	0·13	9·44	0·79

* Values may be too high, due to the accumulation of spray residues on leaf surfaces.

The main points are: (1) There is a close correlation between the percentage of plants affected and tissue test results for Mg. The Mg treatments are reflected by higher values in the plants and the superiority of a foliage spray is shown. Towards the end of the growth period 43 per cent. of the plants were affected in the Nil plots and the Mg tissue value is at L−. (2) The two chemical methods accord in showing high values for Mg in the foliage after spray treatments, low values in the control and soil liquid treatment, and intermediate values for soil treatments. The chemical methods also show that the Mg level in sprayed plants is very high and that the K level is lower than in other plants.

Centre C.

Experiment 1. The effects of various forms of potash on black currant var. Seabrook and red currant var. Laxton's No. 1 have been studied on this plot since 1938. Tissue test and ash analysis data are included in Table IV, together with a record of visual symptoms for the two crops.

The marginal scorch in black currant and more prominently in red currant leaves may result from either a deficiency of potash or an excess of chloride. Tissue tests are of value in distinguishing the two factors responsible for these similar symptoms.

The chloride damage is confirmed in the NP KCl treatment by high tissue test and ash analysis values for Cl, especially in red currant. In the NP (Omit K) plants both chemical methods show low values for K thus confirming the visual signs of the deficiency whereas the Cl levels are normal. The higher Mg and Ca values in the NP (Omit K) plants are shown by the two chemical methods.

Experiment 2. Details of the treatments, visual symptoms, tissue test and ash analysis data are given in Tables V and VI, for apple, Lane's Prince Albert. Symptoms of Mg deficiency appeared first in 1932 but their occurrence has varied with season, being particularly marked in 1939 (4, 5). Plots B_2, C_2, D_1, E_2 and F_1 received a soil application of $MgSO_4$ at 4 cwt. per acre annually from 1940 to 1943.

Spraying treatments, 1944. In 1944 soil applications were discontinued and the following spraying programme introduced :

Plots D_2 (a), sprayed with 1 per cent. solution of $MgSO_4$ (20 per cent. MgO) with sulphonated lorol as spreader.

Plots D_2 (b), sprayed with 2 per cent. solution of $MgSO_4$ with spreader.

The first spray in both treatments was applied at petal fall and further sprays were given at fortnightly periods, making a total of four sprays on May 23rd, June 15th, July 7th and 31st respectively.

Spraying treatments, 1945. During 1945 certain trees in plots E_1, B_2 and D_2 were sprayed with a 2 per cent. $MgSO_4$ solution plus a spreader. Spraying started at petal fall and continued at fortnightly intervals making a total of six sprayings, viz. May 7th, May 22nd, June 9th, June 22nd, July 5th and July 24th. In addition, a few trees in plots A_1 and F_1 were sprayed with K_2SO_4 at 2 per cent. strength.

The main points arising from Tables V and VI are as follows :

Potassium. Symptoms observed in A_2, F_1 and F_2 correspond to M— and L— tissue results and 1·01 to 1·18 per cent. K_2O. In the K spray treatment the K levels are at M+ and total values at 1·36 to 1·41 per cent. K_2O and the absence of visual signs corroborates this result.

Magnesium. The deficiency observed in treatments B_1, E_1 and D_2 are confirmed by tissue test data (L and L+) and total results at 0·20 to 0·25 per cent. MgO. D_2 (low N) is the worst affected and values at L and 0·2 per cent. MgO are in accord with the severity of the visual signs. The Mg spray treatment has raised Mg levels

TABLE IV.

A comparison between visual symptoms, tissue test and ash analysis data for black currant and red currant. Sampling date: 17.8.44.

Treatment.	Crop.	Visual symptoms.	Tissue test data.					Ash analysis as % dry matter.				
			K.	Mg.	Ca.	PO_4.	Cl.	K_2O.	MgO.	CaO.	P_2O_5.	Cl.
NP,K as sulphate.	Black currant.	Nil	H–	M	H	H	M	2·02	0·39	4·26	0·60	0·59
	Red currant.	Nil	H	M+	H	H	M+	2·36	0·46	3·71	0·71	0·62
NP,K as chloride.	Black currant.	Cl toxicity, as trace; pale brown marginal scorch.	H–	M+	H	H	H+	2·26	0·48	4·35	0·63	0·76
	Red currant.	Cl toxicity, severe; as marginal scorch.	H–	M+	H	H	H++	2·33	0·51	3·90	0·67	1·16
NP (Omit K).	Black currant.	K deficiency medium to severe; dark brown marginal scorch.	L–	H	H+	H	M	0·43	0·65	6·21	0·69	0·51
	Red currant.	K deficiency medium.	L–	H	H+	H	M+	0·52	0·70	4·95	0·61	0·61

TABLE V.

The relation between visual symptoms, full chemical data and tissue test results on Lane's Prince Albert apple under various manurial treatments. Season 1944. Sampling date: 27.8.44.

Plot.	Manurial treatment.*	Visual deficiency symptoms.	Chemical data on leaves as percentage dry matter.				Tissue test data on petioles.			
			CaO.	MgO.	K_2O.	P_2O_5.	Ca.	Mg.	K.	PO_4.
A1 ..	Nil	Nil	2·33	0·35	1·58	0·51	M+	M	M	M+
A2 ..		K scorch (medium)	2·92	0·42	0·95	0·75	H—	M	L+	H—
B1 ..	Complete	Nil	2·19	0·29	2·15	0·44	M+	M	H—	M+
B2 ..	Inorganic	Nil	2·38	0·30	1·48	0·48	M+	M	M	M
C1 ..	F.Y.M.	Nil	2·15	0·33	1·74	0·44	M+	M	H—	M+
C2 ..		Nil	2·24	0·40	1·48	0·48	M+	M	M	M
D1 ..	PK (Omit N)	Mg trace	2·04	0·28	2·18	0·82	M+	M—	H—	M+
D2 (a)	1% foliage spray	Mg medium prior to spraying	2·30	0·38	1·96	0·61	M+	M	H—	M+
D2 (b)	2% foliage spray	,, ,,	2·27	0·43	1·72	0·62	M+	M	H—	M+
E1 ..	NK (Omit P)	Mg trace	2·08	0·26	2·20	0·48	M+	M—	H—	M
E2 ..		Nil	2·21	0·30	1·76	0·53	M+	M	H—	M
F1 ..	NP (Omit K)	K scorch slight	2·35	0·33	1·17	0·45	M+	M	L+	M
F2 ..		K scorch trace	2·27	0·30	1·32	0·47	M+	M	M—	M

* Rates of application per acre: F.Y.M., 10 tons; nitrate of soda, 2 cwt.; superphosphate, 3 cwt.; muriate of potash, 1 cwt.

Table VI.

The relation between visual symptoms, full chemical data and tissue test results on Lane's Prince Albert apple under various manurial treatments. Season 1945. Sampling date: 26.8.45.

Plot.	Manurial treatment.‡	Visual deficiency symptoms.	Chemical data on leaves as percentage dry matter.				Tissue test data on petioles.			
			CaO.	MgO.	K_2O.	P_2O_5.	Ca.	Mg.	K.	PO_4.
A1 ..	Nil	Nil	2·52	0·36	1·41	0·51	M	M	M	M−
A2 ..	Nil	K scorch (trace)	2·62	0·40	1·08	0·56	H	M	L+	H−
A2* ..	Spray	Nil	2·65	0·34	1·36	0·63	H	M	M	H−
B1 ..	NPK	Mg trace	2·24	0·25	1·97	0·58	H−	L+	H	H−
B2 ..		Nil	2·41	0·40	1·39	0·51	H	M+	M+	H−
B2† ..		Nil	2·32	0·60	1·48	0·54	H	H−	M+	H−
C1 ..	F.Y.M.	Nil	2·10	0·35	1·70	0·53	H−	M	H−	H−
C2 ..		Nil	2·30	0·43	1·40	0·56	H	M+	M+	H−
D1 ..	PK (Omit N)	Mg trace slight	2·04	0·25	2·10	0·52	H−	L+	H	H−
D2 ..		Mg medium, much defoliation	2·36	0·20	1·63	0·61	H	L	M+	H
D2† ..		Nil	2·32	0·52	1·68	0·56	H	H−	M+	H−
E1 ..	NK (Omit P)	Mg trace, P trace	2·10	0·27	1·98	0·42	H	L+	H	M−
E1† ..		P trace	2·32	0·46	1·70	0·38	H	M+	H−	M−
E2 ..		Nil	2·26	0·34	1·78	0·46	H	M	H−	M−
F1 ..	NP (Omit K)	K scorch (slight)	2·50	0·32	1·18	0·52	H	M	M−	H−
F1* ..		Nil	2·49	0·36	1·66	0·48	H	M	M+	M+
F2 ..		K scorch (medium)	2·43	0·30	1·01	0·56	H	M−	L+	H−

* 2 per cent. K_2SO_4 foliage spray applied. † 2 per cent. $MgSO_4$ foliage spray applied. ‡ Rates of application per acre as for 1944.

and visual symptoms are absent. Despite the non-appearance of symptoms in the presence of potash deficiency, both chemical methods suggest that the Mg values are on the threshold of deficiency. Thus Mg deficiency can be masked when K is also deficient hence the value of chemical diagnostic methods. The threshold of deficiency may be referred to approximately 0·35 per cent. MgO in dry matter or L+ to M— tissue test values.

The spraying method appears to be more effective than soil application in the correction of this deficiency.

Calcium. Full chemical and tissue test results suggest that the levels are adequate throughout. This is in agreement with the presence of 2 to 5 per cent. free lime in the surface soil.

Phosphorus. Both methods confirm that the phosphorus status is lower in the NK (Omit P) treatments and, in 1945, values at 0·38 per cent. P_2O_5 (total) and M— (10 p.p.m.) soluble are at the threshold of deficiency and visual signs of the deficiency, viz. dull purple tints, were recorded.

Experiment 3. The manurial treatments have been previously given. The special point of interest arising from these is the fact that plots A to F have received a complete NPK dressing whereas plots G to L have only received N, and plot Q throughout has received no manure. As a result of these treatments there is a difference in level of K between the NPK plots and the others.

The chemical data for the two varieties Edward VII and Allington Pippin included in the experiment, are shown separately in Tables VII and VIII, for seasons 1944 and 1945.

The main feature, as regards potassium, is that leaves from plots A to F have a higher extractable and total potash content than those from the Omit K treatments.

Var. Edward VII. The leaves from plot Q have 1·2 and 1·3 per cent. K_2O in 1944 and 1945 respectively with a M— tissue test value. Later in the season visual signs of K deficiency were observed in these plots. The total and soluble content in G to L inclusive are near the critical level in 1944 and visual signs were observed in early September whereas in 1945, when chemical results are higher, no symptoms were noted. The threshold of deficiency level approximates to 1·2 per cent. K_2O which corresponds with L+ to M— (25-50 p.p.m. K) tissue values.

Var. Allington Pippin. In this series K deficiency symptoms were first observed in the Q plots when total values are between 1·16 and 1·20 per cent. K_2O and tissue test at L+ (20 p.p.m. K). Although no visual signs of K deficiency were noted in G-H (Omit K plots) the two chemical methods suggest that values are near the threshold of deficiency. The 1·2 per cent. K_2O and L+ 20 p.p.m. total and soluble respectively, suggested as critical values for Edward, are also applicable to the Allington.

As regards magnesium, the values are unsatisfactory throughout and are near or within the threshold of deficiency level.

Var. Edward VII. Visual signs of Mg deficiency in 1944 are more marked when potash is applied, as in A to F, and chemical results L+ to M— (soluble) and 0·22 to 0·34 per cent. MgO (total), corroborate this observation. In 1945 no symptoms of Mg deficiency were noted in the G-L plots and the Mg results at 0·38-0·47 per cent. MgO are higher than in other plots. These values, however, are still near to the critical level. In the Omit K treatments, Mg and K deficiencies occur together and it is difficult to segregate the two by visual inspection. Tissue test and ash analysis confirm that when K is low, Mg is also at the deficient point and when the former is at a high level, Mg deficiency is accentuated. When the two visual signs occur together in the same tree, even in the same leaves, the two chemical methods are invaluable as a diagnostic aid.

Var. Allington Pippin. The soluble and total values are low in plots A-L during 1944, when symptoms were observed in these plots. During the 1945 season symptoms of Mg deficiency were noted in A-F only and this appears to be accentuated by K manuring. In 1944 external signs were noted in K+L with a value of 0·36 per cent. MgO, whereas a similar value for I+J the following season was not accompanied by symptoms. In fixing threshold of deficiency levels for diagnostic work, a marginal range allowing for seasonal variations is better than absolute values.

Centres D, E, F and G.

Experiments on the control of Mg deficiency at these centres provided a further opportunity of testing the use of chemical tissue tests for diagnostic work. The centres are widely separated and site, climate, soil type, manuring and cultural treatment vary considerably. Data are included in Table IX.

The main points are as follows:

Centre D. During 1944 medium to severe symptoms corresponded to low chemical values, viz. 0·10 per cent and L—, total and soluble results respectively. The injection treatment was not clearly reflected probably due to the fact that the leaves sampled were taken from non-injected branches. The foliage spray treatments applied twice in 1944 and three times in 1945 were more effective in reducing the incidence of the deficiency and in increasing Mg levels in the tissues, than the soil treatments. All chemical results suggest that the deficiency has not been fully corrected at this centre.

Centre E. A 2 per cent. foliage spray applied three times during seasons 1944 and 1945 has reduced the effect of the deficiency. Chemical data for Mg are higher in treated leaves than in untreated.

Centre F. The severity markings are confirmed by chemical results, e.g. 0·13 per cent. MgO, and L— (1 p.p.m.) total and soluble respectively, in the non-treated leaves. The soil treatment is reflected by a minor increase in MgO content in both seasons. The 2 per cent. $MgSO_4$ foliage spray treatment applied three times during 1945 has increased values to 0·35 per cent. MgO (total) and M+, 5 p.p.m. Mg (soluble). No leaf symptoms were observed in 1945, yet chemical results as compared with previous seasons, are within the threshold of deficiency. The non-development of symptoms may be due to late frost with subsequent loss of fruit at this centre.

Centre G. Both chemical methods confirm the severity of symptoms. A similar frost effect is evident in 1945 as for centre D, when no visual signs of Mg deficiency were observed. Both chemical methods, although recording higher values than in previous seasons, suggest that the tissue levels are within the threshold of deficiency, in the Nil and soil-treated plots. Lower values appear to be tolerated under such conditions, probably because the loss of fruit results in no translocation of magnesium from the leaves to the fruit.

The potash levels at all centres are high throughout and the effect of Mg deficiency appears to be accentuated.

TABLE VII.

Relation between visual symptoms, full chemical and tissue test data on Edward VII apple under various manurial treatments: 1944 and 1945.

Plot.	Manurial treatment.	Visual symptoms.	7·9·44.				Visual symptoms.	9·9·45.			
			As % dry matter.		Tissue test data.			As % dry matter.		Tissue test data.	
			MgO.	K_2O.	Mg.	K.		MgO.	K_2O.	Mg.	K.
Q	Nil	Nil at this stage but K scorch later	0·33	1·21	M–	M–	Nil, K later in season.	0·37	1·30	M–	M–
A+B	NPK (Spring Application)	Mg slight	0·30	2·0	M–	M+	Mg very slight	0·30	2·32	M–	H–
C+D	NPK (Summer Application)	Mg slight	0·22	2·20	L+	M+	Mg very slight	0·27	2·29	L+	H
E+F	NPK (Autumn Application)	Mg very slight	0·34	1·95	M	M+	Mg slight	0·25	2·16	L+	H
G+H	N (Spring Application)	Mg trace	0·38	1·32	M	L+	Nil	0·43	1·52	M	M
I+J	N (Spring Application)	Mg trace, K trace	0·24	1·46	M–	M–	Nil	0·47	1·50	M	M
K+L	N (Spring Application)	Mg trace	0·37	1·68	M	M–	Nil	0·38	1·62	M	M

TABLE VIII.

Relation between visual symptoms, full chemical and tissue test data on Allington Pippin apple under various manurial treatments: 1944 and 1945.

			7.9.44.					9.9.45.			
			As % dry matter		Tissue test data.			As % dry matter.		Tissue test data.	
Plot.	Manurial treatment.	Visual symptoms.	MgO.	K_2O.	Mg.	K.	Visual symptoms.	MgO.	K_2O.	Mg.	K.
Q	Nil	K trace	0·42	1·23	M	L+	K trace	0·46	1·16	M+	L+
A+B	NPK (Spring Application)	Mg trace	0·23	2·52	L+	M+	Mg trace	0·28	2·36	L+	H
C+D	NPK (Summer Application)	Mg trace	0·20	2·49	M−	M+	Mg trace	0·30	2·52	M−	H
E+F	NPK (Autumn Application)	Mg trace	0·26	2·32	M	M+	Mg trace	0·29	2·40	M−	H
G+H	N (Spring Application)	Mg trace	0·33	1·46	M	L+	Nil	0·39	1·42	M	M−
I+J	N (Spring Application)	Mg trace	0·21	1·56	L+	M−	Nil	0·36	1·50	M	M−
K+L	N (Spring Application)	Mg trace	0·36	1·65	M	M	Nil	0·38	1·46	M	M−

Table IX.

The relation between visual diagnosis, full chemical and tissue test results for Cox's Orange Pippin (Centres D, E, F) and Miller's Seedling (Centre G) under various magnesium treatments for seasons 1944 and 1945.

Centre treatment.	Visual data, severity of Mg deficiency.	Chemical analysis of leaves as % dry matter.		Tissue test data.		Visual data, severity of Mg deficiency	Chemical analysis of leaves as % dry matter.		Tissue test data.	
		MgO.	K_2O.	Mg.	K.		MgO.	K_2O.	Mg.	K.
Centre D. No $MgSO_4$	Medium—Severe	0·11	2·00	L	M+	Severe	0·09	1·96	L	M+
Tree injection with $MgSO_4$ in 1943	Medium—Severe	0·10	1·96	L–	M+	—	—	—	—	—
$MgSO_4$ (soil) and 2% foliage spray	Slight—Medium	0·17	2·23	L+	H–	Trace	0·30	1·70	M	H–
Centre E. No $MgSO_4$	Severe marked defoliation	0·08	2·16	L–	H–	Medium	0·10	2·16	L	M+
MgSO4 2% foliage spray	Slight	0·27	2·20	M–	H–	Very slight	0·34	2·07	M–	H
Centre F. No $MgSO_4$	Medium—Severe	0·13	3·56	L–	H–	(Frost damage)	0·20	2·20	L+	H
$MgSO_4$ soil application	Medium	0·20	2·64	L+	H–	,,	0·25	2·13	M–	H
2% foliage spray 3 times in 1945	,,	0·20	2·64	L+	H–	,,	0·35	2·06	M	H
Centre G. (*Miller's Seedling*) No $MgSO_4$	Severe	0·10	2·92	L	H–	(Frost damage)	0·20	1·97	M–	H
$MgSO_4$ (soil application)	Medium—Severe	0·15	2·67	L+	H–	,,	0·32	1·89	M	H
$MgSO_4$ (soil) and 2% spray	,,	0·15	2·67	L+	H–	,,	0·51	2·06	M+	H
$MgCO_3$ (soil treatment)	,,	0·18	2·29	L+	H–	,,	0·31	1·99	M–	H

DISCUSSION.

The tissue test method has been used to diagnose the mineral status of potato, tomato, black currant and apple grown at centres differing widely in soil characters.

The results show that tissue test standards may be fixed which are characteristic of the nutrient and crop and that mineral deficiencies or toxicities can be determined with reference to these standards.

Tissue tests proved of value as a complementary method to visual diagnosis as is illustrated by (1) the acidity complex in potato at centre A, where K, Mg, Ca, P and N deficiencies and Mn toxicity may occur together and symptoms may be difficult to interpret by visual means alone ; (2) K deficiency and Cl toxicity in red currant at centre C, Experiment 1, since both nutrient disorders produce a marginal scorch ; (3) K and Mg deficiencies in apples ; in some varieties, e.g. King Edward VII (centre C, Experiment 3) both may produce marginal scorch effects. In such instances chemical methods are necessary to provide confirmatory data and the tissue test is convenient.

At deficiency and toxicity levels there is a close correlation between the tissue test results and those of ash analysis. Thus it was possible to confirm K, Mg, Ca, P and N deficiencies and Mn and Cl toxicities using the two methods. Although the tissue test data are less precise than those of ash analysis the fact that several determinations can be made in a short time tends to offset that disadvantage.

SUMMARY.

1. The tissue test method has been applied to the diagnosis of K, Mg, Ca, P and N deficiencies and Mn and Cl toxicities in potato, tomato, black and red currant and apple, grown at widely different centres.

2. The results reflected the manurial treatments, whether applied to the soil or as foliage sprays.

3. The tissue test data showed good agreement with the visual method and this test is of particular value when visual symptoms are difficult to interpret. This was demonstrated in the following complex nutrient disorders : (*a*) acidity complex in potato where K, Mg, Ca, P and N deficiencies and Mn toxicity occurred together ; (*b*) K deficiency and Cl toxicity in black and red currant ; (*c*) K and Mg deficiencies in apples.

4. There was a close correlation between tissue test and ash analysis data over the diagnostic range for deficiencies and toxicities, for all crops studied. The comparisons suggest that the tissue test method has a wide application in the study of mineral disorders in plants.

ACKNOWLEDGMENTS.

The work described in this paper was financed by grants made by the Agricultural Research Council under the special scheme of the Council for the investigation of mineral deficiencies in crops. Thanks are due to the Council for its generous support of the work and to Professor T. Wallace for his advice and interest during the course of this investigation.

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(*Received* 23/2/48.)

STUDIES IN BIENNIAL BEARING

III. GROWTH STUDIES IN "ON" AND "OFF" YEAR TREES

By L. B. SINGH

East Malling Research Station, Kent

Introduction.

In the second paper of this series (Singh, 1948) a review of the literature on biennial bearing was made, and the conclusion was drawn that detailed studies of the various aspects of growth in "on" and "off" year trees might help to show the fundamental causes of the trouble. Accordingly a series of studies was designed, in which the following factors were investigated, in relation to "on" and "off" year condition.

A. Tree vigour, as shown by (1) shoot growth, (2) trunk thickening, and (3) root growth.

B. Shoot development, i.e. time and rate of growth of shoots and their leaves.

C. Spur development, (1) in relation to a biennial bearing variety in "on" and "off" years, and (2) in different varieties of contrasting regular and biennial habits.

In addition to these, the material available provided opportunities, which were taken, for studies of the relation of "on" and "off" year conditions to :

D. Leaf colour and leaf scorching, and,

E. Winter injury to shoots. (This last is being reported elsewhere.)

Material.

With the exception of the root growth studies and the studies on regular and biennial bearing varieties, of which the material will be described later, all the work was carried out on bush trees of Miller's Seedling apple, a variety noted for its biennial bearing habit. The trees were approximately thirty years old at the beginning of the experiments and had shown the biennial tendency for many years. Individual tree records of crop yields were available from 1937, when the trees came under the control of the Research Station. A summary of the cropping of the thirty-six experimental trees, from 1937 to 1944, during the whole of which period they had received uniform treatments, is shown in Fig. 1, which shows, at a glance, the general biennial tendency of cropping. This is confirmed by the figures for the factors "B" and "I" calculated by the method of Hoblyn *et al.* (1936), which show that "B" (the regularity of biennial fluctuation) = 83·33, and "I" (the intensity of fluctuation) = 0·78 over the period. The possible maximum of "B" is 100, and this would have been obtained in this series, if the 1937 crop (which was destroyed by frost) had exceeded that of 1938. The maximum for "I" is 1·0, and this is only obtained where the crop in alternate years is nil. Thus, it can be seen that this series of trees had a very marked and intense biennial habit.*

Following the severe spring frost of 1944, all of these trees were entirely " in step " in 1945, and blossomed profusely. For the present biennial bearing experiments it was desirable to have comparable trees both in " on " and " off " year condition in the same season. Nine trees were therefore entirely deblossomed by hand in April, 1945. This placed them in " off " year condition so far as crop was concerned and had the effect of making them blossom fully in 1946 when the rest of the trees were in their " off " year. Thus in 1946 when the present writer began his investigations nine trees in " on " year condition and twenty-seven in " off " year condition were available, providing excellent material for studying the behaviour of trees in both phases of the biennial rhythm, under the same climatic conditions.

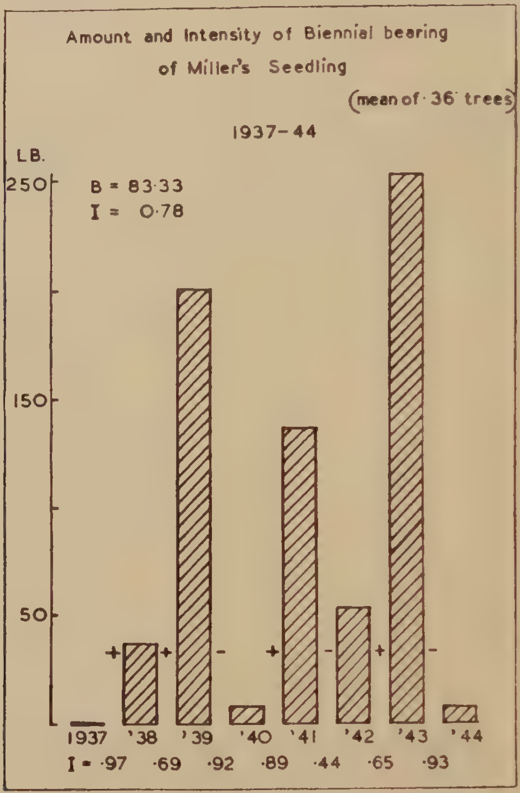

FIG. 1.

$$* \; B = \frac{\text{number of pairs of differences of unlike sign between successive crops}}{\text{total number of pairs of differences}} \times 100$$

$$I = \frac{\text{difference between successive crops}}{\text{sum of successive crops.}}$$

The methods of calculation are more fully reviewed in Paper II (Singh, 1948).

For the growth studies, two sets of four comparable trees were selected in 1946; Series A in "on" and Series B in "off" year condition. Two trees from each series were used for intensive records on sample leaders and spurs, which are described more fully later in this paper.

A. TREE VIGOUR, IN RELATION TO BIENNIAL BEARING.

1. *Shoot growth.* The number and length of all the one-year-old shoots over 5 cm. long on the trees of Series A and B were recorded in the winters of 1945-46 and 1946-47, separate records being made of leaders and laterals.* Thus data were obtained for the shoot growth in the season 1945, when the trees of Series A were in an artificial or "abnormal off year" condition and those of Series B in a normal "on" year condition, and for season 1946, when the trees of Series A and B were in normal "on" and "off" year conditions respectively. The number, length and average length of shoots are summarized in Table I.

TABLE I.

Miller's Seedling apple. Amount of shoot growth in "on" and "off" year trees. (Total for four trees.)

1945.	SERIES A. Phase, "On" Year. Av. crop 382 lb.			SERIES B.* Phase, "Abnormal Off Year". Av. crop 6 lb.			Significance P=0·05.		
	No.	Length. cm.	Av. length cm.	No.	Length. cm.	Av. length cm.	No.	Length.	Av. length.
Leaders	157	3,543	22·6	377	9,342	24·7	Sig.	Sig.	n.s.
Laterals	1,006	13,246	13·2	2,890	53,556	18·5	Sig.	n.s.	Sig.
Both	1,163	16,789	14·4	3,267	62,898	19·2	Sig.	Sig.	n.s.
1946.	Phase, "Off" Year. Av. crop 0 lb.			Phase, "On" Year. Av. crop 306 lb.					
	No.	Length. cm.	Av. length. cm.	No.	Length. cm.	Av. length. cm.	No.	Length.	Av. length.
Leaders	113	3,907	34·6	135	5,457	40·4	n.s.	n.s.	n.s.
Laterals	1,869	42,113	30·4	2,093	51,618	24·7	n.s.	n.s.	n.s.
Both	1,982	46,020	30·7	2,228	57,075	25·6	n.s.	n.s.	n.s.

* "On" year trees completely deblossomed in April, 1945.

Table I shows that the Series B trees, i.e. those which had been deblossomed, and thus were in an "abnormal off year", produced very much more growth than those of Series A, the normal "on" year trees. The number and length of shoots was more than twice as much in the former as in the latter, and the difference was visible both in leaders and laterals. All the differences in number and length are statistically significant, except the length of laterals, in which, on account of variability of material, the difference did not quite reach the level of significance.

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* Leaders are the apical shoots of each branch; laterals, the side shoots.

In this case, however, the average length of lateral shoots was significantly greater in the deblossomed trees than in the others.

A different state of affairs is seen in 1946, however, when the trees of Series A were in a normal " off " year and those of Series B in a normal " on " year condition. In this year, the normal " off " year trees produced slightly less growth than the normal " on " year trees ; but none of the differences is significant. Actually the trees of Series B were, on the average, slightly larger than those of Series A, and allowing for this would give almost similar figures for the " on " and " off " year trees. Thus it appears that the length of shoot growth of trees in normal " on " and " off " year condition is not strikingly different ; but that shoot growth is markedly stimulated by the act of deblossoming an " on " year tree.

Pruning weight and shoot thickness. The one-year-old shoots of 1945 and 1946 were pruned by the normal spur pruning method, and a record of their weight was obtained. A representative sample of 250 cm. length of pruned shoots was weighed to give an estimate of shoot thickness. The diameter of base of shoots of small samples of leaders and laterals was also measured. The results are given in Table II.

TABLE II.

Miller's Seedling apple. Weight and thickness of pruned shoots in " on " and " off " year trees. (Total for four trees.)

1945.	SERIES A. Phase, " On " Year. Av. crop 382 lb.	SERIES B. Phase, " Abnormal Off Year ". Av. crop 6 lb.	Significant difference. P=0·05.
Weight pruned shoots—gm. ..	2,098·0	10,824·0	4,432·0
Weight 250 cm. length pruned shoots—gm.	29·3	53·5	6·42
Mean diameter at base of leaders —mm.	3·69	5·21	0·20
Laterals—mm.	3·65	5·46	0·13
1946.	Phase, " Off " Year. Av. crop 0 lb.	Phase, " On " Year. Av. crop 306 lb.	
Weight pruned shoots—gm. ..	8,685·0	8,477·0	n.s.
Weight 250 cm. length pruned shoots—gm.	58·1	41·4	n.s.
Mean diameter at base of leaders —mm.	4·61	5·42	n.s.
Laterals—mm.	4·46	5·13	n.s.

The figures given in Table II confirm those of Table I and show a still greater contrast in 1945. The weight of pruned shoots of the trees of Series B in 1945 was more than four times that of the trees of Series A. This increase of contrast, over that shown in Table I, arises from the fact that shoots of Series B were also thicker than those of Series A, and it can be seen in Table II that the differences, both in weight per unit length and in mean diameter of the base of the shoot, are significant.

Once more, in 1946, no significant difference appeared either in total weight or in thickness of shoots. It is interesting to note that although the total length of shoots in 1946, shown in Table I, is smaller in Series A than in Series B, the total weight shown in Table II is greater. The mean weight per unit length of shoot is nearly 50 per cent. greater but did not reach statistical significance.

It should be emphasized that only one year's comparable records are here available for the contrasts between the normal " on " and " off " year trees (1946), and between deblossomed and fruiting " on " year trees (1945). The contrasts in the latter case were so marked that it may reasonably be concluded that similar differences would be seen in other cases where the blossom of " on " year trees is destroyed. In the case of normal " on " and " off " year trees the differences are not well marked, and it is clear that further observations on a larger number of trees over a series of years are needed to establish what, if any, are the real differences.

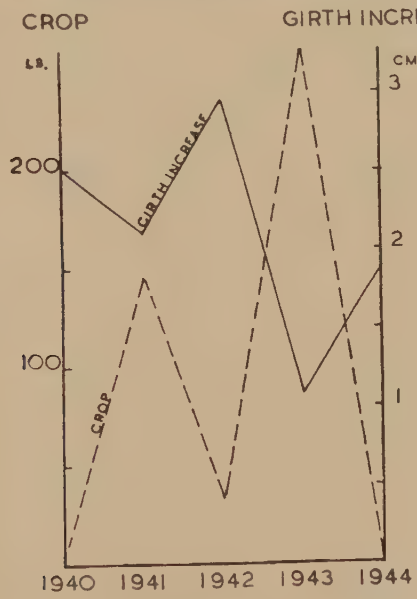

FIG. 2.

Relation between crop and girth increase. Miller's Seedling. 1940-44.

2. *Trunk thickening.* The mean trunk girth increase and crops of eighteen of the uniformly treated Miller's Seedling trees are shown in Fig. 2.

It is clear from this graph that the girth increase is negatively related to crop yield. Similar records on the trees of Series A and B (four trees of each) for 1945 and 1946 showed that the mean girth increase in the deblossomed trees of Series A in 1945 was 3·40 cm. while in the " on " year trees of Series B it was only 1·52 cm. In 1946 their mean girth increases were 0·68 cm. and 1·53 cm. respectively. Thus these data confirm those shown in Fig. 2, and suggest that the developing fruits make a first call on the available food materials and take priority over trunk thickening.

3. *Root growth.* In the root-growth studies of Rogers (1939) some data on relation of root growth to cropping of mature Lane's Prince Albert apples, for a period of four years, are available. The paper quoted was primarily a study of the

week-to-week variation in root growth, as seen in glass-walled observation trenches; and although it was noted that in a year when frost destroyed the whole crop, root growth was greatly increased, no detailed comparison of amount of crop and root growth was made. The full figures have been made available to the present author through the kindness of Dr. W. S. Rogers. The year-to-year comparison of root growth and crop is shown in Fig. 3.

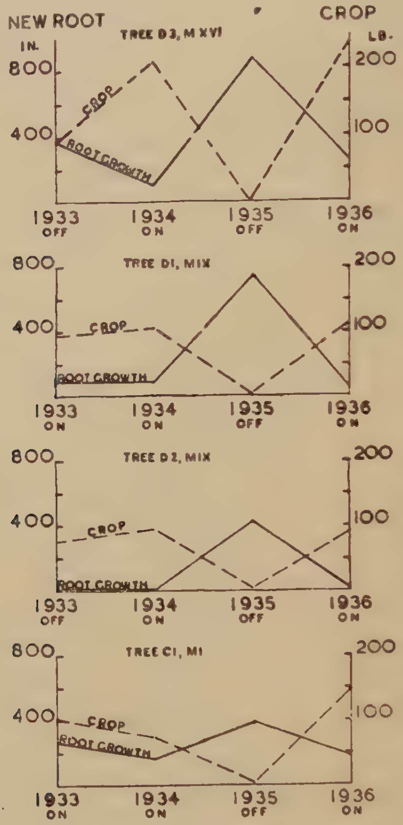

FIG. 3.

Root growth of Lane's Prince Albert on various rootstocks in relation to their annual crop yield.

Fig. 3 shows clearly that in the case of all four trees studied there is a close negative relationship between the amount of crop and root growth. It is also clear that the relationship is more marked in the "off" year which resulted from frost in 1935. The addition of new roots in that year has been relatively more than in any normal "off" year, and similar results may be expected when a tree is prevented from bearing a crop by artificial means, e.g. complete bud-rubbing or deblossoming.

General conclusions from the studies on tree vigour. These studies have shown clear negative relationships between heavy cropping and both amount of trunk thickening and amount of new root growth. The relationship between heavy cropping and number, length and thickness of new shoots is less clear. So called " abnormal off year " conditions, caused by blossom removal, appear to stimulate growth of shoots and trunk to a greater extent than normal " off " year conditions. Thus a distinction needs to be drawn between conditions existing when trees, which would normally have cropped, are put into an " off " year condition by a frost or similar cause and trees which are in a normal " off " year condition following a heavy crop. In the former case the tree is in a more vigorous condition, because the absence of crop in the previous year has allowed it to store greater reserves and moreover to increase its absorbing and synthesizing organs. In the latter case the growth of the crop, which is entirely a nutrient-consuming process, has left the tree relatively weaker, and also restricted the production of roots.

Thus it may be inferred that the tree is in a more vigorous condition at the beginning of a normal " on " year than at the beginning of a normal " off " year, but this extra vigour is all used up by the developing fruits. The important effects upon the leaves are dealt with later.

The fact that the shoot growth differs relatively little in normal " on " and " off " year trees, while the trunk thickening and root growth both differ greatly, suggests that the shoots, being nearest to the leaves, make the first demand on food materials, but that the developing fruit takes priority over stem thickening and root growth.

B. SHOOT DEVELOPMENT.

1. *Time and rate of growth of shoots.* Although, as has been shown in the previous section, shoots from normal " on " and " off " year trees differ relatively little in their total extension length, there may, as suggested by Finch (1935) be important differences in period and rate of growth. Such differences might affect the production and utilization of food reserves of the trees at various critical periods and thus possibly affect the formation of fruit buds by spurs. Serial observations on sample shoots and spurs from " on " and " off " year trees were therefore made over a period of two years.

Material and Methods. Four trees of Miller's Seedling, two in the " on " and two in the " off " year (Series A and B respectively), as described in Section A, were chosen for the serial observations. Four branches, two on the north and two on the south side of each tree, were selected ; and on them ten leaders and ten laterals were labelled for weekly recording. The laterals measured in this experiment were those arising from the bud nearest to the leader.

Measurements from the base of the shoots to the base of the bunched-up leaves which cover the growing point were made, to the nearest millimetre, at weekly intervals all through the shoot growth period in 1946 and 1947.

Observations. In 1946 shoot growth began about April 28th, on both " on " and " off " year trees. Apical buds were the first to move, and grew faster than lateral buds. During the first three weeks there was not much difference between growth rate of " on " and " off " year trees, though the " on " year trees grew slightly faster (Fig. 4).

In a second stage, from about the end of May to the middle of July, a marked contrast appeared. In this stage the " on " year shoots, both leaders and laterals, grew nearly twice as fast as the " off " year ones. In the third stage from mid-July to mid-August, the " off " year shoots grew much faster than " on " year ones.

The " off " year leaders nearly caught up the " on " year ones in length, and the " off " year laterals actually exceeded the " on " year ones. In the final stage, from mid-August onwards, the rate of growth slowed down and finally ceased. However, even in this period the " off " year shoots grew relatively faster than the " on " year ones.

The 1947 results, shown in Fig. 5, are similar to those of 1946 for the first two periods, but different thereafter. Shoot growth in 1947 started a week later than in 1946, possibly due to the severe winter of 1946-47. The first stage continued until about the beginning of June. The second was similar to that of 1946 ; but the third stage was less marked in the leaders and practically absent in the laterals ; and shoot growth left off sooner than in 1946. This appears to be mainly due to the very dry summer of 1947. Fig. 6 shows that rainfall was plentiful in August, 1946, and soil moisture tension (as recorded by a Rogers' tensiometer) was low during the latter part of August and September. This undoubtedly increased shoot growth in this period. In Fig. 7 it can be seen that there was practically no rain from the third week in July until mid-September. Soil moisture tension remained above the maximum reading of the instrument from about August 9th to September 30th. This notable drought certainly checked shoot growth. It may be that other factors also contributed to the contrast in the growth rates, but the effect of soil moisture appears to be outstanding.

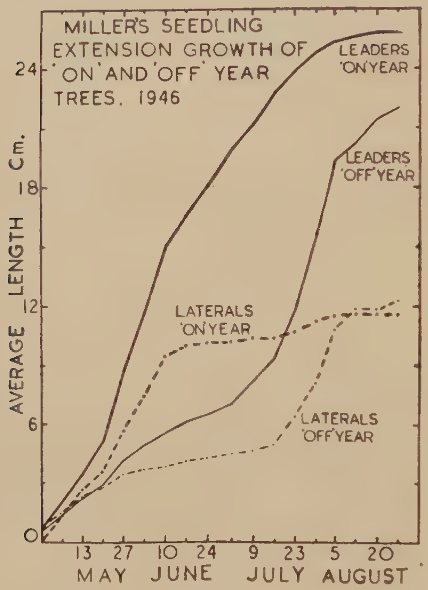

FIG 4.

Shoot growth cycle of " on " and " off " year trees, 1946.

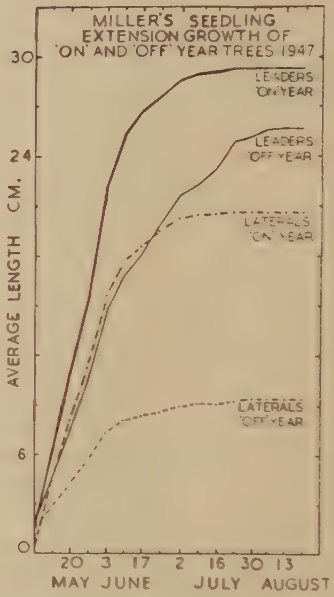

Fig. 5.

Shoot growth cycle of " on " and " off " year trees, 1947.

Discussion. It is clear from these results that the periodic growth of shoots differs in " on " and " off " year trees under similar climatic conditions, thus the seasonal march of shoot growth, though also dependent on normal climatic factors such as temperature and moisture, is greatly influenced by the physiological condition of the tree, as expressed in the "on" and "off" year phases.

The number of shoots on these mature trees is relatively small in relation to the number of spurs on the whole tree, i.e. about 500 shoots and 4,000 spurs per tree, but nevertheless these shoots may well have great effects on the food supplies and reserves of the tree and thus possibly on the differentiation of fruit buds. In this connection it may be worth while to test the following hypotheses.

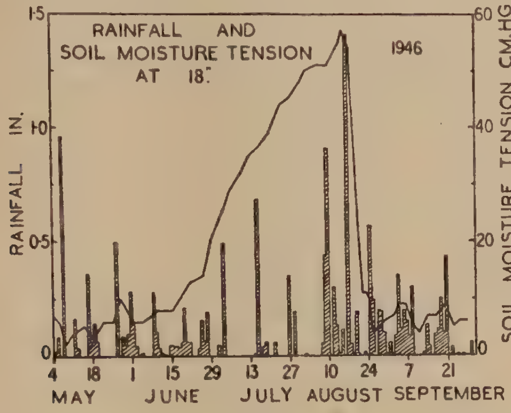

FIG. 6.

Rainfall and soil moisture tension at 18 in. deep, 3 ft. from the trunk of one of the experimental trees, 1946.

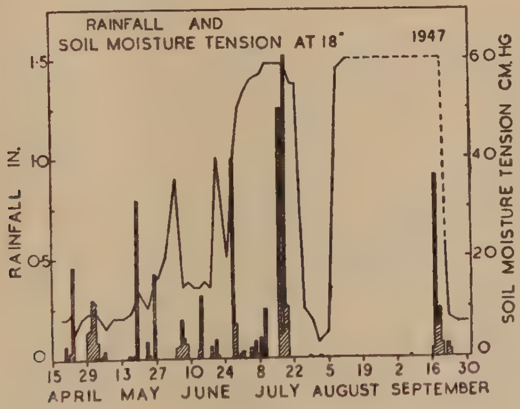

FIG. 7.

Rainfall and soil moisture tension, 1947. The dotted line shows the period in which tension exceeded the upper limit of the instrument.

(i) The most critical period, conditions in which are likely to influence the differentiation of fruit buds, is generally accepted to lie in May and June. The present work shows that during this period, shoot growth in "on" year trees is rapid, whereas that of "off" year ones is relatively slow. It is possible that the rapid shoot growth of "on" year trees puts a heavy demand on the food supply of the tree throughout the critical period of fruit-bud differentiation so that a great portion of it goes to meet that demand and therefore very little is left available for fruit-bud differentiation; and hence very few or no fruit buds are formed. Further, these "on" year trees have also to maintain a very heavy crop for the current season. Fruits start growing and swelling from about the beginning of June, which may further restrict the existing supply and thus aid in checking fruit-bud differentiation. A relationship between shoot and fruit growth during the critical period of fruit-bud formation would be worth investigating. In the case of "off" year trees, the conditions are just the reverse and hence a sufficient number of fruit buds is formed.

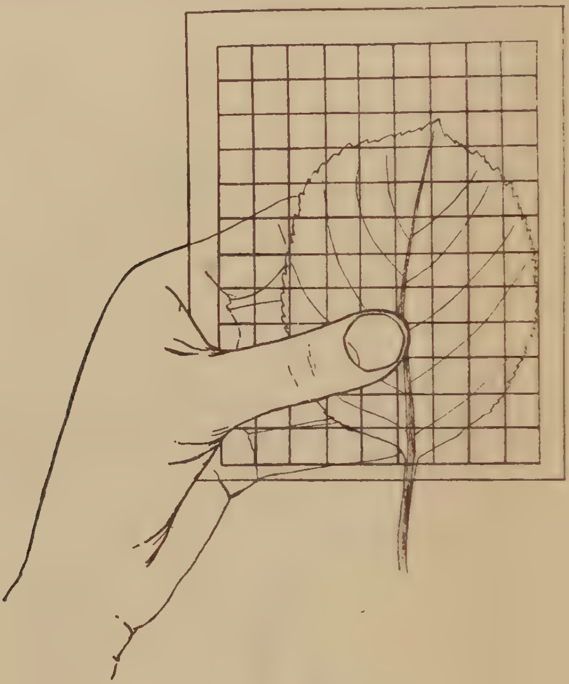

FIG. 8.

Method of using leaf area integrator.
No. of complete squares = 38.
No. of incomplete squares = 7 complete squares.
Area of leaf = 45 sq. cm.

(ii) It is also possible, according to Loeb's hypothesis, that rapidly growing shoots in the "on" year may be manufacturing an increased amount of substances which possess fruit-bud inhibitory effects and consequently check their differentiation whereas in "off" year trees, manufacture of such substances is likely to be restricted by slow growth and so there is no interference with fruit-bud formation.

2. *Leaf growth on shoots.* Although the bulk of the leaves are borne on the spurs (and a detailed study of these is reported below (page 134), the leaves on the shoots also play an important part in the nutrition of the tree. Therefore a study

was made of the leaf development, on the same shoots as those used for the extension growth studies. All unfolded leaves were counted at fortnightly intervals and their area was measured each month.

The leaves were measured on the tree by means of a modified form of the "integrator" described by Vyvyan and Evans (1932). In this modified form, which was suggested by Dr. Rogers, the integrator consisted of a single sheet of celluloid whose surface was divided into squares of 1 centimetre by straight lines ruled with Indian ink. Two sizes were used, 7 × 6 cm. for small leaves and 9 × 12 cm. for larger ones. The advantage of this simplified form was its lightness and ease of handling. To use it, the leaf was lightly supported by the fingers of one hand and the integrator held in place by the thumb so that one border of the leaf coincided with a line in the integrator (Fig. 8). The number of full squares covering the leaf surface was then counted. The contiguous fractions round the margin of the leaf were balanced into complete squares and added to the total number of full squares. If some fraction still remained unmeasured, it was counted as a whole square if it were greater than half a square, and was omitted if less than half. This method, using 1 cm. squares, proved quick and of a very satisfactory degree of accuracy for the relatively large number of leaves involved in the samples. Leaf and extension growth measurements were taken on the same day on both sets of trees. The records for 1946 and 1947 are summarized in Tables III and IV.

TABLE III.

Number of leaves per shoot on sample shoots of Miller's Seedling in "on" and "off" year conditions during two seasons.

	LEADERS.				LATERALS.			
	"On" year.		"Off" year.		"On" year.		"Off" year.	
1946.	Total No. leaves	No. leaves per unit length.	Total No. leaves	No. leaves per unit length.	Total No. leaves	No. leaves per unit length.	Total No. leaves	No. leaves per unit length.
May 13 ..	3·9	1·15	3·5	1·59	3·4	1·26	4·4	1·91
27 ..	6·1	0·69	6·0	1·55	5·5	0·97	5·4	1·59
June 10 ..	10·3	0·69	7·2	1·31	8·5	0·90	7·1	1·87
24 ..	11·8	0·65	9·3	1·40	9·8	0·97	8·3	1·98
July 9 ..	14·8	0·69	11·4	1·39	10·0	0·98	8·3	1·80
23 ..	15·2	0·64	12·7	1·09	10·7	1·00	10·4	1·62
Aug. 5 ..	16·4	0·65	16·4	0·85	11·8	1·02	12·8	1·18
12 ..	16·4	0·64	16·4	0·81	12·0	1·03	13·5	1·14
1947.	"On" year.		"Off" year.		"On" year.		"Off" year.	
May 13 ..	7·2	1·22	7·8	1·73	6·3	1·43	6·3	2·30
27 ..	9·4	0·60	10·0	0·96	7·9	0·70	7·8	1·39
June 10 ..	13·4	0·53	11·5	0·68	10·6	0·60	7·9	0·98
24 ..	14·9	0·54	13·6	0·69	12·1	0·62	8·2	0·95
July 9 ..	15·0	0·52	14·1	0·63	12·3	0·60	9·0	1·00
23 ..	15·0	0·51	14·7	0·59	12·3	0·60	9·0	1·00
Aug. 5 ..	15·0	0·51	15·7	0·61	12·3	0·60	9·0	1·00
12 ..	15·0	0·51	15·8	0·62	12·3	0·60	9·0	1·00

Results. It may be seen in Table III that the total number of leaves per leader or per lateral was similar in " on " and " off " year trees in both years ; but the " on " year trees tended to have more leaves than the " off " year ones in early summer, following the same general lines as the shoot extension growth, already discussed. The " off " year shoots had a relatively greater number of leaves *per unit length,* however, than " on " year ones, all through the shoots' growth period.

TABLE IV.

Area of leaves per shoot on sample shoots of Miller's Seedling in " on " and " off " year conditions, during two seasons.

1946.	LEADERS.				LATERALS.			
	" On " year.		" Off " year.		" On " year.		" Off " year.	
	Total area leaves in sq. cm.	Area leaves per unit length in sq. cm.	Total area leaves in sq. cm.	Area leaves per unit length in sq. cm.	Total area leaves in sq. cm.	Area leaves per unit length in sq. cm.	Total area leaves in sq. cm.	Area leaves per unit length in sq. cm.
June 10 ..	145·8	9·72	121·3	22·05	90·3	9·61	61·6	16·21
July 9 ..	307·1	14·49	208·5	25·43	182·9	17·76	145·1	32·98
Aug. 5 ..	379·3	14·99	347·6	18·01	208·9	18·17	188·6	21·43
1947.	" On " year.		" Off " year.		" On " year.		" Off " year ".	
June 10 ..	278·6	11·0	303·0	18·0	199·3	11·3	190·9	23·6
July 9 ..	282·0	9·7	345·6	15·5	201·4	9·8	207·0	22·7
Aug. 5 ..	282·0	9·6	345·6	13·5	201·4	9·8	207·0	22·0

Table IV shows that total leaf area on leaders and laterals was rather greater in the " on " year than in the " off " year trees in 1946, but rather less in 1947. A consistent contrast is seen, however, in the area of leaves *per unit length of shoot*, which was greater in " off " year shoots than that in " on " year ones both in 1946 and 1947.

It is apparent therefore that there is more leaf growth in the " off " year than in the " on " year on the extension shoots when measured in relation to their length, thus supporting other evidence that relatively more food is available for other purposes in " off " year than in " on " year trees.

C. SPUR DEVELOPMENT.

In Miller's Seedling, and indeed in most apples, most of the blossom buds are borne on the spurs, which constitute the main fruiting organs of the tree. A detailed study was therefore made of the spur behaviour of trees of the biennial bearing variety Miller's Seedling on the one hand and of various regular and biennial bearing varieties on the other, including records of their development, at regular intervals, for two years.

1. *Spur behaviour of Miller's Seedling in " on " and " off " years.* It is convenient to deal with this study under the following headings:

(*a*) Blossoming and fruiting performance of spurs.
(*b*) Leaf growth of spurs.
(*c*) The life history of spurs as shown in serial photographs.

Material and method. The same trees were used as in the shoot development studies previously described. Eighty spurs on the " on " year trees (Series A) and 80 on the " off " year ones (Series B), distributed along branches on the north and south sides of the trees, were labelled and numbered for serial observations in the spring of 1946. The recorded data were: time of growth; nature of growth, i.e. shoots or clusters; presence or absence of blossom; presence or absence of fruits; number of unfolded leaves; area of unfolded leaves and period of leaf fall.

The records of leaf number and area were taken on only 40 simple (i.e. single) spurs, 20 " on " year and 20 " off " year, as it was impracticable to record more.

For the photographic study, complex spurs (i.e. spur systems) consisting of three or more simple spurs, were used. The spur systems of these mature trees usually contained at least five or six simple spurs.

Each fruit bud of Miller's Seedling usually produces five blossoms, all of which usually set fruits. In all the spurs under experimental treatment the fruits were thinned to one per spur about a fortnight after setting.

(*a*) *Spur blossoming and fruiting performance.* A summary of the blossoming and fruiting performance of the 160 marked spurs is given in Table V.

This shows, in the first place, that the trees of both series behaved in an entirely biennial manner in both years, irrespective of the climatic differences already mentioned (page 130). The factor " I ", based on the number of fruits set on the selected spurs, is 0·97 and 1·00 for the two sets of spurs (or Biennial Index 97 and 100).

Twenty-three spurs (15 per cent. of the total), ten in Series A and thirteen in Series B, remained vegetative in both years. The remaining 137 spurs (85 per cent. of the total) blossomed in one year or the other. Of these, 126 spurs (92 per cent.) set fruit and only eleven spurs (8 per cent.) did not. It was noted that all the spurs which blossomed but did not set fruit did not produce blossom again in the following year. Only one spur (in Series A) was out of step with the rest. None of the sample spurs blossomed in both years.

Thus, of the 160 spurs observed, 23 (15 per cent.) failed to fruit in both years because they produced no blossom. Eleven (6 per cent.) failed to fruit because the bloom did not set, and the remaining 137 (79 per cent.) blossomed and set fruit in alternate years. It is therefore clear that, in Miller's Seedling, the biennial habit is almost entirely due not to the failure of blossom to set fruit, but to the failure of individual spurs to produce blossom in consecutive years.

(*b*) *Leaf growth of spurs.* This study proved to be one of the most important of the series. Leaf growth was recorded fortnightly in 1946 on forty spurs, ten on each of the two " on " year and two " off " year trees. The spurs recorded in the " off " year were all vegetative (i.e. they produced no blossom) whereas those in the " on " year were all fruiting. Records were repeated on the same spurs (apart from the replacement of three damaged ones) in 1947, when the former vegetative spurs had become the fruiting ones and vice versa. All the unfolded leaves (including those from bourse buds, as discussed later) were counted and their area measured, fortnightly, as previously described.

TABLE V.

Blossoming and fruiting performance of spurs of Miller's Seedling.

Tree series.	No. spurs selected.	1946.				1947.				
		Cropping phase.	No. spurs blossoming.	No. spurs setting fruit.	No. spurs vegetative.	Cropping phase.	No. spurs blossoming.	No. spurs setting fruit.	No. spurs vegetative.	Factor "I"*
A.	80	"On"	69	62	11	"Off"	1	1	79	0·97
B.	80	"Off"	0	0	80	"On"	67	63	13	1·00

* Based on number of spurs setting fruit.

The mean number and area of leaves per spur is shown in Fig. 9. Considering number, it is clear that both in 1946 and 1947 the " on " year trees had rather more leaves than the " off " year ones at the beginning and end of the season and less leaves in the middle of the season. The initial increase continued until the end of May in both years. From mid-June till the beginning of autumn defoliation, " off " year trees had more leaves than " on " year ones, owing to the shedding of a number of primary leaves by the " on " year trees (as discussed later). This contrast was less marked in 1947 than in 1946, possibly because of the dry weather in 1947. A third contrast visible is that the spurs on " off " year trees defoliated more rapidly than the " on " year ones in both years. This defoliation usually occurred in an acropetal order. These contrasts, although consistent and interesting, are much smaller than the contrasts in leaf area which are shown in the lower part of the figure.

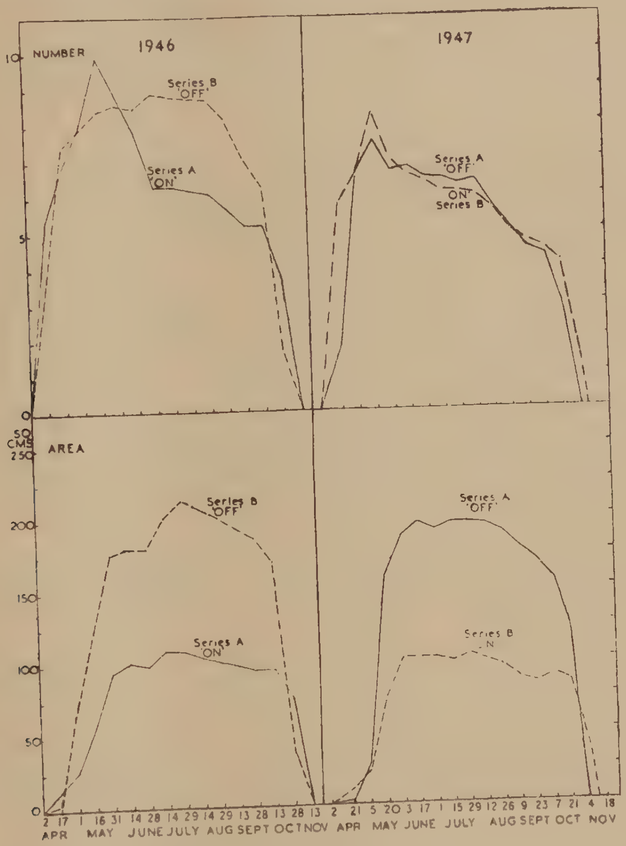

FIG. 9.

Number and area of leaves on spurs in " on " and " off " phases for two successive years.

It is seen that in the first fortnight of April, " on " year trees have a larger leaf area than " off " year ones, owing to earlier leaf production. From then onwards, however, leaf area on " off " year spurs increased much more rapidly than the " on " year ones and remained much greater, in fact nearly twice as much, throughout the season until the main defoliation period.

Since the difference in number of leaves is relatively small, it is clear that increase in leaf area on the " off " year spurs was due to the larger size of the individual spur leaves. The mean size of a fully expanded spur leaf at the end of July on " off " year trees was 24·3 sq. cm. in 1946 and 31·1 sq. cm. in 1947, while that of " on " year trees was only 17·2 sq. cm. and 16·9 sq. cm. in 1946 and 1947 respectively.

In both years the autumn defoliation of " off " year spurs started earlier and proceeded more rapidly than the " on " year ones. It was interesting to note that in 1947 defoliation of the spurs started earlier than usual, probably owing to the severe drought, but was subsequently checked for some weeks, following heavy rain, and recommenced at the normal time.

The most important point shown by these graphs is that in the period between May and July, in which fruit bud differentiation occurs, the " off " year trees have a much greater leaf area per spur than the " on " year ones.

(*c*) *Life history of spurs in " on " and " off " years.* The records obtained in Sections (*a*) and (*b*) contain detailed data on certain important points, but do not give a complete picture of the biennial cycle of growth. In order to give a clearer view of the characteristic behaviour of spurs in the " on " and " off " year, serial photographs of sample spurs were taken at important stages of growth (normally every month) for two years. The photographs were all made under field conditions, with a quarter-plate camera. On account of the need for economy in illustrations, only a selection of the photographs, which show the main points, are here presented.

The life cycle of the spurs may conveniently be dealt with in various main stages as under :

(i) *Pre-blossom stage* (*April*). Both types of spurs were similar in appearance before bud-break except that the fruit buds of " on " year spurs were bigger and fatter (Fig. 10) than the leaf buds of " off " year spurs (Fig. 11). A few weeks later the fruit buds gave rise to the typical pink blossom clusters (Fig. 12) whereas the leaf buds unfolded only leaves (Fig. 13). From then onwards their behaviour differed considerably as described below.

(ii) *Blossoming and fruiting stage* (*May to August*). *" On " year trees.* The leaves of the fruiting spurs of these trees increased rapidly for a fortnight after full bloom and relatively slowly thereafter. This check corresponded with the onset of fruit swelling. At this time secondary buds, below the blossom cluster base, began to grow. Most of these developed leaf clusters, but a few developed shoots. These are termed " bourse bud leaves " and " bourse bud shoots " (Swarbrick, 1928). The percentage of bourse bud shoots was found to be very small. Out of 69 fruiting spurs recorded (in two years) only seven developed bourse bud shoots and 62 produced bourse leaf clusters as seen in Fig. 16 and Fig. 20, Spurs 7 and 8. Figs. 20 and 21, which show Spur B in its " on " phase in the previous year, are included both as good pictures of bourse bud production and as illustrations of the change of phase.

The first few primary leaves below a blossom cluster are quite small, about 4 to 6 sq. cm. in area, but the later ones are larger (Fig. 14). The bourse bud leaves are much bigger than the primary leaves (see Fig. 20, Spur 8). It is interesting to note also that the size of both primary and bourse bud leaves on the fruiting spurs remained much smaller throughout the season than those on vegetative spurs, as already discussed in Section (*b*) above.

When the bourse bud leaves were about a month old, about the middle of June, the primary leaves on the fruiting spurs began to fall, and in a few weeks most of the fruiting spurs had lost practically all of them and possessed only bourse bud leaves or no leaves at all. The loss of leaves at this critical time was probably of great significance, as this was the period when the fruit buds were expected to differentiate for the following year. This period is also of great importance in the growth of the current season's fruit. Fruits on those spurs which had lost all the primary leaves, and had also failed to produce bourse bud clusters, made no further increase in size. The size and development of the bourse bud leaves and the scars left by the fall of primary leaves are visible in Fig. 21 taken after the fruit was harvested.

" *Off* " *year trees.* In the meantime, the spurs of the " off " year trees, which were all vegetative and produced no flowers, exhibited quite different features. The primary basal leaves persisted throughout the season till the natural autumnal defoliation. No bourse bud shoots or leaves were produced. The primary leaves grew much more rapidly than those of fruiting spurs and in fact they doubled their size between the middle and end of May (Fig. 15).

A few of these spurs did not settle down as leaf clusters but developed into shoots (Fig. 17, right) ; the proportion of such spurs was, however, very small (two out of 80). These shoots continued growing and producing new leaves throughout August, whereas the remaining spurs had settled down after unrolling their normal number of primary leaves (Fig. 17, left).

(iii) *Post fruiting and defoliation stage* (*September to November*). During the first part of this period the fruiting spurs had markedly darker green leaves than the vegetative ones. During this stage the vegetative spurs produced relatively large central buds which appeared as if they would blossom in the next season (Fig. 19) whereas the fruiting spurs formed small and slender buds (Fig. 18). Defoliation took place in November, and from November to the following April the buds remained dormant.

The illustrations given are mainly 1947 photographs, as they were technically a better series, but exactly similar descriptions applied to the 1946 series, when the spurs observed were in their opposite phases and gave an exactly opposite behaviour. Two examples of Spur B in the " on " phase in 1946 are given in Figs. 20 and 21.

Thus the description of the two-year cycle of spur behaviour of the biennial bearing variety Miller's Seedling is exactly the description of typical " on " year behaviour followed by typical " off " year behaviour, or vice versa.

Two full series of photographs, each covering two years, are given in the thesis of which this paper formed a part. Spur A, shown here, actually belonged to a different experiment (on the same plot), but has been chosen for reproduction as it provided some of the best photographs of typical " on " year behaviour.

2. *Spur behaviour of regular and biennial bearing varieties in successive years.* This formed a complementary study to the " on " and " off " year trees described above, and included four varieties, Miller's Seedling and Blenheim Orange representing biennial bearers, and Ellison's Orange and Ribston Pippin as regular bearers. This selection was also made so that each set included one diploid and one triploid variety. Miller's Seedling and Ellison's Orange are diploids and Blenheim Orange and Ribston Pippin triploids. Eight trees of each of these four varieties, which had received

similar cultural treatment in the past, were chosen for recording. The trees weer cordons on M.IX, approximately 18 years old. During the studies, the usual routine summer pruning was omitted as it would have interfered with the growth observations: but winter pruning was carried out.

(*a*) *Blossoming and fruiting performance of spurs.* Forty simple spurs from each variety, i.e. five spurs, distributed over the length of the cordon, on each of eight trees, were labelled early in the spring of 1946. Fruits which set on these spurs were thinned to singles, as on the rest of the trees. The blossoming and fruiting behaviour of these spurs over a period of two years is shown in Table VI and a summary of the regularity of individual spur performance is given in Table VII.

Considering first the two diploid varieties (Miller's Seedling and Ellison's Orange), Table VII shows that 83 per cent. of the spurs of Miller's Seedling were biennial in blossoming. The remaining 17 per cent. remained vegetative in both years. On the other hand 73 per cent. of the total spurs on Ellison's Orange blossomed regularly in both years; of the remaining spurs, 22 per cent. blossomed in alternate years and 5 per cent. were vegetative in both years.

The details of number of spurs setting and maturing fruits in each year are given in Table VI. In both varieties, those spurs which blossomed set fruits heavily (three to four fruitlets per truss) and in both cases fruit drop was relatively light; but it was rather heavier in Ellison's Orange (14 per cent. in 1946 and 20 per cent. in 1947) than in Miller's Seedling (3 per cent. only).

The factor " I " based on number of spurs setting fruits is 1·00 for Miller's Seedling and only 0·09 for Ellison's Orange. The factor " I " based on number of fruits harvested rises to 0·23 in the case of Ellison's Orange, due to the higher fruit drop; but on either basis of calculation the contrast between the regular and biennial behaviour respectively of spurs of the two varieties is clearly seen.

Both tables show that the contrast arises in the first place from the biennial or regular blossoming of individual spurs rather than from differences in setting of fruits or in the " June drop ".

The triploid varieties, Blenheim Orange (biennial) and Ribston Pippin (regular), gave similar contrasts in spur behaviour. Table VII shows that 50 per cent. of the spurs of Blenheim Orange blossomed in alternate years only. The remaining 50 per cent. were vegetative in both years.

On the other hand, 68 per cent. of the spurs of Ribston Pippin blossomed in both years and all the remaining spurs (32 per cent.) blossomed in alternate years only. None of these spurs remained vegetative in both years.

The fruit set, as shown in Table VI, was fairly heavy on both these varieties, though not quite so heavy as in the case of Miller's Seedling and Ellison's Orange. In Blenheim Orange 75 per cent. of the blossoming spurs set fruits and in Ribston Pippin 88 per cent. In both varieties the " June drop " of fruitlets was relatively high; in Ribston Pippin 49 per cent. of the set fruitlets in 1946 and 79 per cent. in 1947 were shed by the end of the first week in June. Thus very few spurs carried fruits to maturity. On Blenheim Orange also, 47 per cent. of the spurs that set fruits shed them early in June.

The factor " I " based on number of spurs setting fruit is 1·00 for Blenheim Orange and 0·19 for Ribston Pippin, which again shows the great contrast in spur behaviour between the regular and biennial varieties. The high fruit drop in Ribston Pippin causes the factor "I", based on the fruit harvested, to rise to 0·60; while it

TABLE VI.

Blossoming and fruiting performance of spurs of regular and biennial bearing varieties.

Variety.	Cropping phase.	No. spurs selected.	No. spurs flowering.	No. spurs setting fruit.	No. fruits shed early in June.	No. spurs vegetative.	No. fruits matured.	Factor " I ".	
								Based on spurs setting fruit.	Based on fruits matured.
Miller's Seedling (biennial)	1946—" Off "	40	0	0	0	40	0	1·00	1·00
	1947—" On "	40	33	32	1	7	27		
Ellison's Orange (regular)	1946—" On "	40	35	35	5	5	10	0·09	0·23
	1947—" On "	40	29	29	6	9	16		
Blenheim Orange (biennial)	1946—" Off "	40	2	0	0	38	0	1·00	1·00
	1947—" On "	40	18	15	7	22	5		
Ribston Pippin (regular)	1946—" On "	40	40	35	17	3	12	0·19	0·60
	1947—" On "	40	27	24	19	12	3		

Table VII.

Summary of regularity of blossoming of individual spurs of regular and biennial bearing varieties.

Variety.	Total.	No. of spurs.			% of spurs.		
		Blossoming in both years.	Blossoming in alternate years.	Vegetative in both years.	Blossoming in both years.	Blossoming in alternate years.	Vegetative in both years.
Miller's Seedling (biennial)	40	0	33	7	0	83	17
Ellison's Orange (regular)	40	29	9	2	73	22	5
Blenheim Orange (biennial)	40	0	20	20	0	50	50
Ribston Pippin (regular)	40	27	13	0	68	32	0

remains 1·00 for Blenheim Orange. The number of fruits matured is too small, however, for a good calculation of " I ", so this factor was also calculated for the total crop of the eight trees of each variety, in the two years. This calculation gave " I " figures as follows: Miller's Seedling and Blenheim Orange 1·00; Ribston Pippin 0·30; Ellison's Orange 0·05. Thus it is seen that Ribston Pippin is relatively regular in comparison with Miller's Seedling and Blenheim Orange, but is less regular than Ellison's Orange.

It is of interest to consider also whether the spurs which carried fruits to maturity were less likely to do so in the following year than those which shed their fruits. The numbers available are small, but examination of the records shows that in the case of Ellison's Orange, eight of the ten spurs which carried fruits to maturity in 1946 also did so in 1947. In the case of Ribston Pippin, all three of the spurs which carried fruits to maturity in 1947 had also done so in 1946. Thus it is clear that in these varieties, the carrying of fruits to maturity does not necessarily preclude the spurs from doing so in the next season.

To summarize, it is clear that nearly 70 per cent. of the spurs on Ellison's Orange and on Ribston Pippin produced blossoms and set fruit regularly in both years; while in contrast, none of the spurs on Miller's Seedling or Blenheim Orange did so. Therefore the classification of the two former as relatively regular and the two latter as biennial is correct.

The " I " figures, both for fruits set and fruits matured, show up a finer gradation, i.e. that Ellison's Orange is more regular than Ribston Pippin.

Blenheim Orange is just as biennial as Miller's Seedling, as in both cases " I " was 1·00; but in addition only 50 per cent. of the spurs of Blenheim Orange blossomed even in the " on " year phase of the biennial cycle, showing that Blenheim Orange is not only biennial but a shy cropping variety as well.

In all the cases quoted it is clear that the biennial bearing habit is dependent mainly on failure of individual spurs to produce blossom in consecutive years. Each variety has, however, shown certain interesting varietal features of spur growth associated with its biennial or regular behaviour, which will be discussed more fully in Section (*b*) below.

(*b*) *Life history of spurs in successive years.* In order to illustrate the contrast in spur development of the two regular and two biennial varieties, serial photographs were made of one complex spur of each variety in the same manner as described above. The main points of interest are as follows:

(i) *Miller's Seedling.* The photographs of the complex spur on the cordon trees of Miller's Seedling showed results exactly comparable with those of the bush Miller's Seedling trees, illustrated in Figs. 10-21. The cordon trees were in their " off " year in 1946, when they had no blossoms or fruits, and in the " on " year in 1947, when they produced blossom profusely, as already described. There is no need therefore to include a second series of photographs and the description already given will serve conveniently for comparison with that of the regular varieties described below.

(ii) *Ribston Pippin.* The main contrasts in spur growth are shown in Figs. 22 to 25, which show the same spur at various stages in 1946 and 1947. Figs. 22 and 25 show that the Ribston Pippin spurs blossomed profusely in both years and the full series of photographs shows similar behaviour in other respects. This is in marked contrast to the widely differing behaviour, in successive years, of spurs of Miller's Seedling.

Figs. 22 and 23 show the blossoms and developing fruits in 1946. In contrast with the Miller's Seedling the primary leaves on the fruiting spurs of the Ribston Pippin did not fall in June, as can be seen in Fig. 23. Another important feature in the spur development of Ribston Pippin is the production of bourse buds which, in contrast to those on the fruiting spurs of Miller's Seedling, formed fruit buds for the following year and thus enabled the same spur to blossom regularly in consecutive years.

Bourse buds, which began developing in June, can be seen at the bases of two of the developing fruits on the right in Fig. 23. The formation of blossom-bearing dards and secondary shoots at the bases of fruits is also a common character in Ribston Pippin. A short dard and a shoot, which has been pruned, are seen on the left in Fig. 24.

In Fig. 25 it is seen that both bourse buds and dard have produced blossom clusters. This, of course, shows that the bourse buds had differentiated into fruit buds in the previous summer.

The blossom clusters, produced on the bourse buds and dards, set fruits but the fruits of this spur were shed in the " June drop ", a prominent character of Ribston Pippin, as already mentioned. The fruiting spurs and the dards again developed plump bourse buds, almost exactly similar to those of April, 1947 (Fig. 24). Thus it appears that the regular blossoming will persist from year to year.

(iii) *Ellison's Orange.* The spur behaviour of Ellison's Orange is generally similar to that of Ribston Pippin. Only two illustrations are therefore given. Fig. 26 shows the profuse production of blossom in 1947 on Spur D, which had blossomed equally well in 1946. The bold primary leaf clusters are a feature. Fig. 27 shows the developing fruits, in August, and three large persisting primary leaves and four bourse bud leaves can be seen at the base of Spur 9. Thus, like Ribston Pippin, Ellison's Orange produced strong bourse buds and dards which fruited in the following season and also produced further strong bourse buds and dards.

(iv) *Blenheim Orange.* This variety, as already stated, is not only biennial but a very shy cropper. It was in the " off " year in 1946 and one of the three selected buds (Spur 3) developed a shoot and the other two (Spurs 4 and 5) vegetative rosettes (Fig. 28). The buds both at the beginning and end of the season were slender.

In the next season it is seen (Figs. 29 to 31) that once again these slender buds produced no blossom, although they should have been in the " on " year in 1947. As already shown in Table VII only 18 out of a total of 40 spurs blossomed even in the alternate years and only five fruits were carried to maturity. No prominent differences in appearance were observed between the spurs which blossomed in alternate years and those which remained consistently vegetative.

It is clear from the photographs and other visual observations that these spurs are not lacking in leaf area in the " off " year. Thus some other factor must be responsible for the shy cropping performance.

Discussion. It is apparent, from the spur behaviour of the varieties studied, that their difference in regular or biennial cropping depends mainly on the behaviour of the bourse buds, formed at the cluster bases of the fruiting spurs. In the regular varieties, the bourse bud differentiates into a fruit bud at the same time as the spur on which it forms is bearing a fruit. In the biennial varieties, the bourse bud which forms on a fruiting spur is apparently too weak to differentiate into a fruit bud during that season and requires a further season, in which it remains as a vegetative cluster, before it becomes a fruit bud.

Further, in the regular varieties, the primary leaves of the fruiting spurs persist throughout the season, and these varieties also produce numerous dards and secondary shoots. In the biennial variety, Miller's Seedling, the primary leaves fall early in the season and practically no dards are formed. Thus it appears that the failure of the bourse buds of Miller's Seedling to become fruit buds may be connected with their relatively smaller number of supporting leaves.

It has been shown above that " off " year trees have a much greater leaf area than " on " year ones and, in the " off " year season, plenty of fruit buds are formed. It appears therefore that it is the lack of leaf surface, at a critical period, on the fruiting spurs of biennial bearing varieties which prevents the differentiation of bourse buds into fruit buds for the following year. On the other hand, bourse buds of the fruiting spurs of regular varieties have enough leaf area, all through the season, to enable them to become fruit buds for the next year and thus to maintain the regular blossoming habit of the variety.

D. LEAF COLOUR AND LEAF SCORCH.

The contrasting performance of trees in " on " and " off " years suggests that their physiological condition is considerably different, and thus visible differences may well be expected in leaf colour and leaf scorch. Filewicz (1935) in Poland, reported that in autumn " on " year trees had leaves of relatively dark colour, and " off " year trees much lighter ones. He considered that the darker green leaves of the " on " year trees were due to the fact that fruit development had used up food supplies and so delayed the maturing of shoots.

Leaf colour records were taken on " on " and " off " year Miller's Seedling trees about the end of August, 1946, when most of the extension shoots had stopped growing and the majority of leaves had fully developed. The leaf colour was assessed by eye, all trees being assessed by the same experienced recorder, and placed in one of the following categories : 1.=yellow green, 2.=light green, 3.=dark green.

Twenty-eight trees were recorded, of which twenty-three were in their " off " year and had no crop and the rest had crops varying from 280 lb. to 400 lb. per tree. All the fruit had been picked and removed by the time the leaf colour record was made.

On examination of the records, it was found that all of the " on " year trees had been placed in category 3, whereas all of the " off " year trees fell into category 2. The differences became even more marked a fortnight later. The contrast was so great that it was easy to identify which trees had borne a crop simply by the darker green colour of their leaves. It may well be that developing fruits had used up a relatively high proportion of the carbohydrates manufactured by the leaves, and thus left the nitrogen content relatively high.

In the second week of October, visual records of marginal and interveinal leaf scorches were made on the same trees. The trees were placed in six categories, as below :

0=no scorch on leaves.
1=1-10 per cent. of leaves showing scorch.
2=11-20 per cent. of leaves showing scorch.
3=21-30 per cent. of leaves showing scorch.
4=31-40 per cent. of leaves showing scorch.
5=41-50 per cent. of leaves showing scorch.

Interveinal scorch was found on only one tree ; but most of the trees, both in " on " and " off " years, showed some marginal scorch. A summary of marginal scorch records gave the mean category value of marginal scorching for the " on " year trees as 3·4, and that for "off" year ones 1·9, i.e. there was much more scorch in " on " year trees than in " off " year ones. This greater tendency to scorching, in trees bearing a heavy crop, has been previously noted by Hatton and Grubb (1924) and is probably due to the demand of the fruits for certain nutrients. It is certainly a clear indication of a difference in the chemical status of leaves in " on " and " off " year trees at the end of the season.

GENERAL DISCUSSION AND CONCLUSIONS.

Studies in tree vigour as expressed by root growth, trunk growth, shoot growth and leaf growth in " on " and " off " year trees showed that the physiological condition of the tree at the beginning of an " on " year is quite different from that at the beginning of an " off " year. This is further evidenced by the greater increase in wood growth produced in an " abnormal off year ", when the blossom of a tree in " on " year condition is prevented from setting fruits.

The leaf growth studies have shown that leaf area is greatly restricted in the presence of heavy crop. Doubtless this not only lessens the amount of carbohydrates available in the individual spurs, but it also is responsible for the reduction in trunk thickening and root growth that has been observed. The reduction of root growth is of double importance, in that it automatically restricts the absorption of nutrients, and thus affects every other growth activity.

There seems little doubt that the extra leaf area available in " off " year spurs is largely responsible for the differentiation of fruit buds upon them. This conclusion is strengthened by the evidence from the bud-rubbing and blossom-thinning trials, in which an increase in leaf area, in relation to fruits produced, resulted in greater differentiation of fruit buds.

The spurs form the main fruiting organ of the trees and it is their behaviour which determines whether the tree will or will not bear a crop. These studies have shown that, although the general nutritional status of the tree differs greatly in " on " and " off " year conditions, the spurs may, to quite a large extent, behave as individuals, for some may blossom while the majority are vegetative, and vice versa.

It is the lack of blossom production which causes the biennial behaviour of varieties such as Miller's Seedling, and these growth studies have shown that this arises from the failure of the bourse buds, produced below the fruiting clusters, to differentiate into fruit buds for the following spring. This appears to be due to lack of sufficient leaf surface during the critical period for fruit bud formation.

The variety Blenheim Orange provides a contrast, however, for this very vigorous variety produced abundant leaves, and yet failed to form many fruit buds, even in the " off " years. This shy cropping as well as biennial behaviour may be due to its varietal make-up, and may also have some connection with the fact that, as a very vigorous triploid variety, it was unsuitable for growing in cordon form.

The regular cropping varieties, Ellison's Orange and Ribston Pippin, on the other hand, formed bourse buds which usually differentiated into fruit buds each year, and this gave them their regular bearing habit. It seems probable that the leaf surface of these spurs, at the critical period, was sufficient to enable differentiation to take place.

It may be noted that the triploid variety, Ribston Pippin, was less regular than the diploid variety, Ellison's Orange.

SUMMARY.

1. A series of intensive studies of the shoot, trunk, spur and leaf development of the typically " biennial " bearing apple variety Miller's Seedling and studies on several other regular and biennial bearing varieties have been carried out, with the object of gaining further information on the causes of the biennial habit.

2. A strong negative correlation between heavy cropping (i.e. " on " year condition) and trunk thickening in Miller's Seedling was found. From data on Lane's Prince Albert apple a similar negative relationship between heavy cropping and root growth was established.

3. No significant differences in shoot growth between trees of Miller's Seedling in normal " on " and " off " year condition were shown ; but it was found that in trees which were artificially put into " off " year condition by de-blossoming in the year following a normal " off " year, significantly more shoot growth was produced. This showed that the physiological condition of a tree at the start of an " on " year is completely different from its condition at the start of an " off " year ; and thus " on " year trees whose blossom is destroyed by frost or similar causes are placed in a condition which is different from that of normal " off " year trees and is here called an " abnormal off year " condition.

4. The extension shoots of " on " year trees start growing sooner and grow faster than those of " off " year trees during the critical period of fruit bud formation (i.e. May and June). It is suggested that this may affect the differentiation of fruit buds in biennial bearing trees.

5. Studies of leaf number and area, throughout the season, on the spurs of " on " and " off " year trees showed that the leaf area per spur of " off " year trees was nearly double that of " on " year trees during the critical period of fruit bud formation. This is considered of great significance.

6. Records of individual spur behaviour showed that, in Miller's Seedling, 80 per cent. of the spurs blossomed, all together, in alternate years only, and the rest remained vegetative. Thus the biennial habit in this variety is due to lack of blossom production in alternate years, not to lack of setting of blossoms that are produced. In Miller's Seedling the bourse buds (secondary buds which form below fruiting clusters) fail to differentiate into fruit buds. This is the main cause of the biennial behaviour of this variety and it appears, from these studies, to be due to lack of leaf surface at the critical period.

7. A comparative study of spur behaviour of two " regular " varieties (Ellison's Orange and Ribston Pippin) and two " biennial " ones (Miller's Seedling and Blenheim Orange) showed that, while Miller's Seedling behaved as described above, Ellison's Orange and Ribston Pippin both produced blossoms regularly each year on more than 70 per cent. of their spurs, and a high proportion of these also set fruits each year. In these two varieties, the bourse buds differentiated into fruit buds each year. Blenheim Orange, however, differed from the rest, for not only did all of its spurs fail to blossom in alternate years, but also even in the " on " years only 50 per cent.

of its spurs blossomed, and relatively few of these set fruits. Thus this variety, besides being biennial in bearing, is a shy cropper.

8. Classing the four varieties according to intensity of fluctuation of crop, Ellison's Orange was the most " regular " cropper, and Ribston Pippin next. Both Miller's Seedling and Blenheim Orange showed the maximum intensity of fluctuation, but, as already stated, Miller's Seedling cropped heavily in the " on " year and Blenheim Orange only lightly.

9. A study of leaf colour and leaf scorching (mainly marginal scorch) showed that " on " year trees had darker green leaves than " off " year ones, towards the end of August. Leaf scorching was more severe in " on " year trees than in " off " year ones.

ACKNOWLEDGMENTS.

The writer wishes to record his warm thanks to Dr. R. G. Hatton, Director of East Malling Research Station, for providing the experimental material and facilities for these studies. He would specially thank Dr. W. S. Rogers, who supervised these studies, for his constant advice, encouragement and help throughout the work. He would also thank Dr. M. C. Vyvyan for helpful advice and Mr. J. Taylor for help with the statistical procedure.

Thanks are also due to Miss F. Bacon for much help with the recording, to Miss M. H. E. Davies and Miss F. Bacon for help with the preparation of diagrams and to Miss E. C. Thompson, Mr. W. F. Bond and Mr. T. Y. Jones for taking the photographs.

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(*Received* 25/2/48.)

PLATE I.

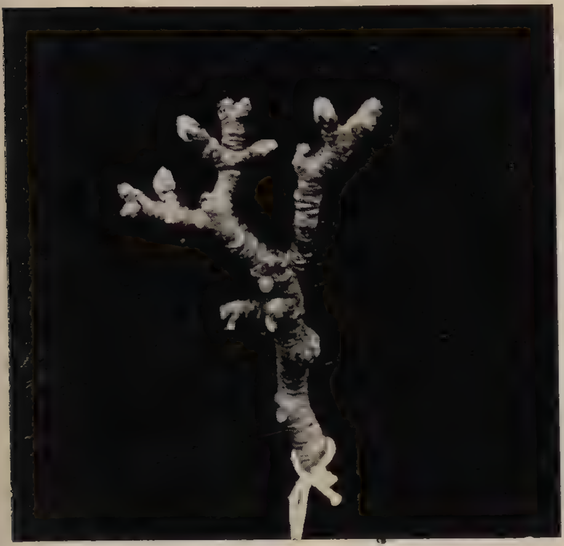

FIG. 10.
Spur A, *Miller's Seedling*, in “on” phase. April 1st, 1947. Note plump fruit buds.

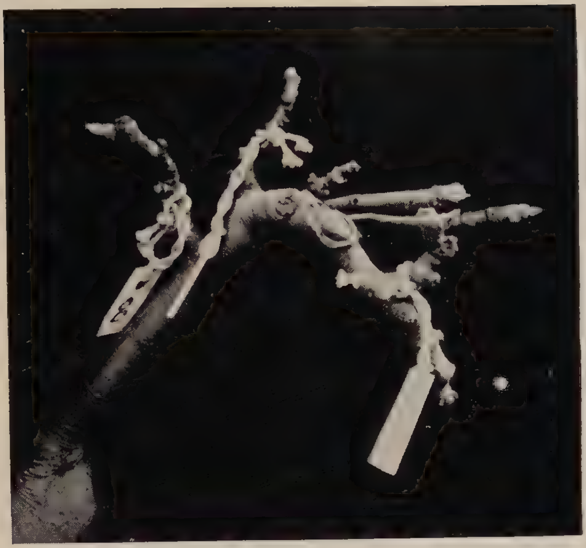

FIG. 11.
Spur B, *Miller's Seedling*, in “off” phase. April 1st, 1947. Note slender vegetative buds.

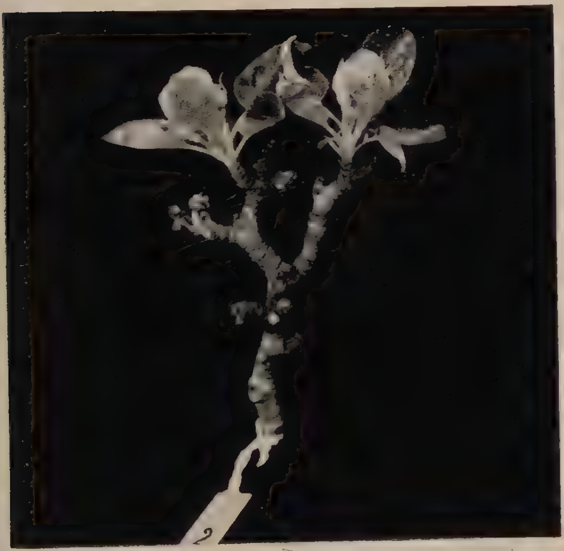

FIG. 12.
Spur A, May 1st, 1947. Blossom opening.

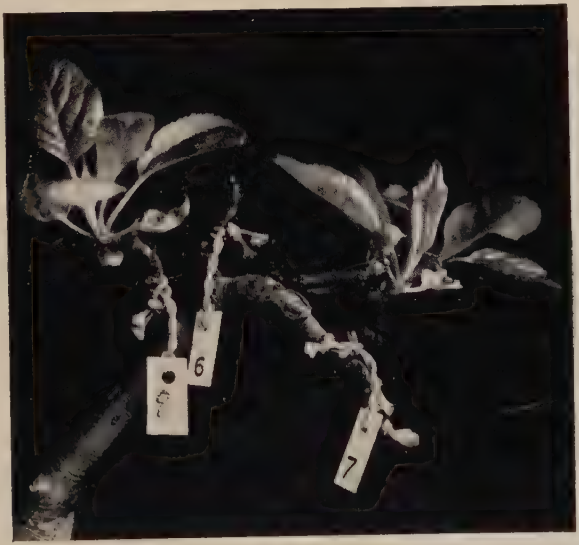

FIG. 13.
Spur B, May 1st, 1947. Vegetative clusters.

PLATE II.

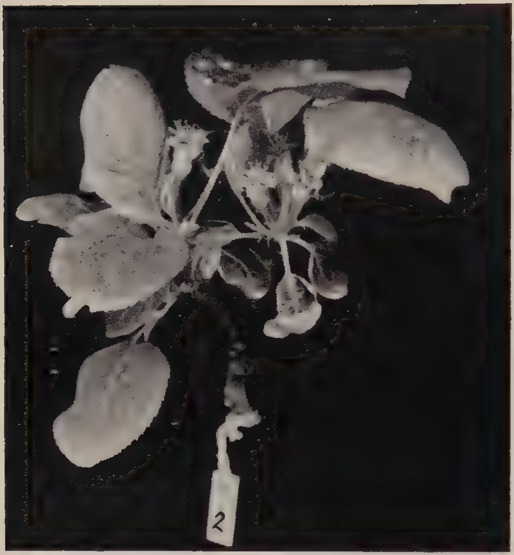

FIG. 14.
Spur A, May 21st, 1947. Note small primary leaves at base of fruitlets.

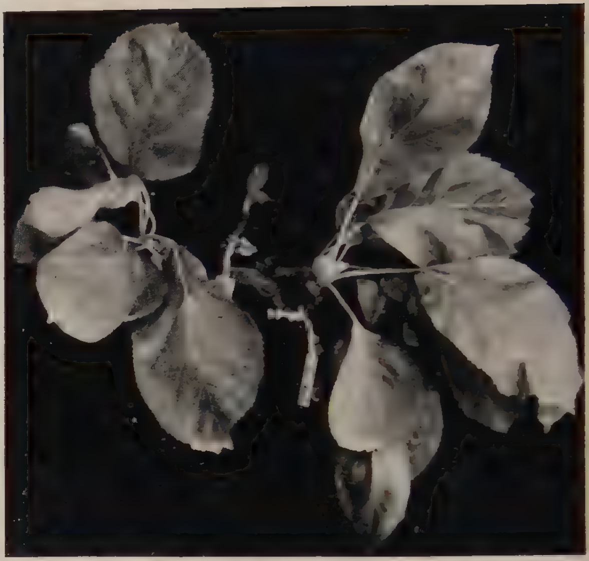

FIG. 15.
Spur B, June 10th, 1947. Note large primary leaf clusters.

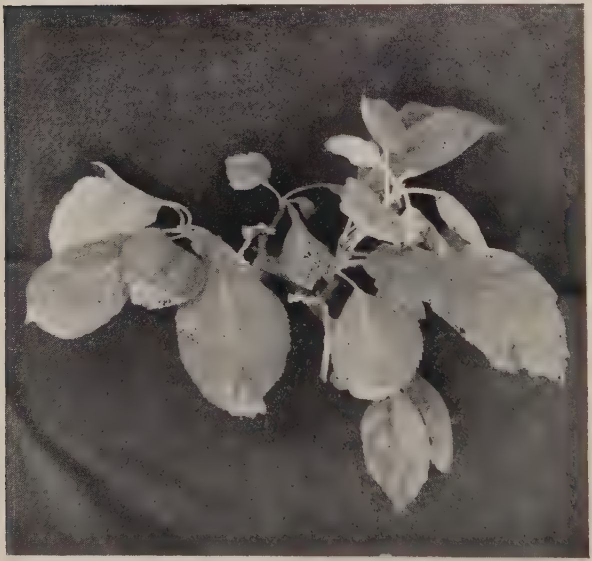

FIG. 16.
Spur A, August 5th, 1947. Note bourse bud cluster at base of fruit.

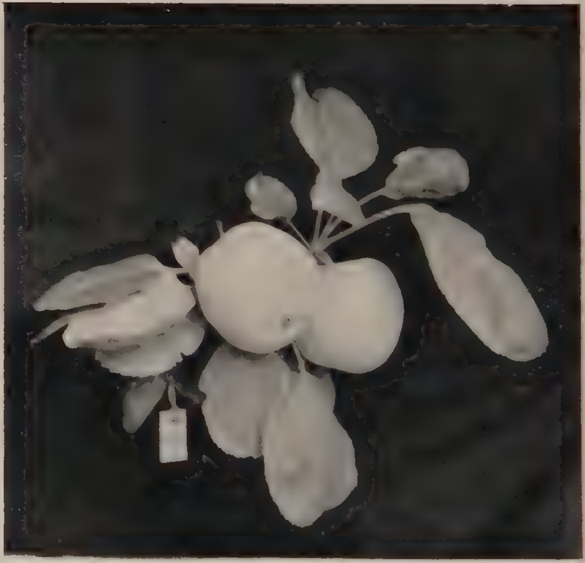

FIG. 17.
Spur B, July 3rd, 1947. Note retention of primary leaves, and production of a dard, on right.

PLATE III.

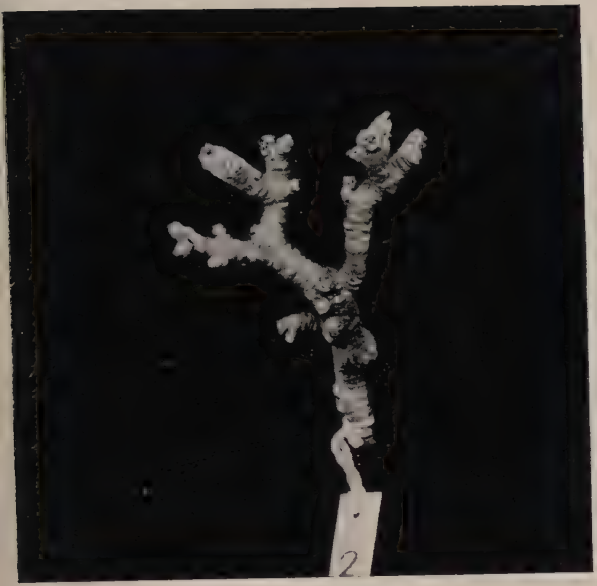

FIG. 18.

Spur A, November 7th, 1947. Note weak bourse buds, compare Fig. 10.

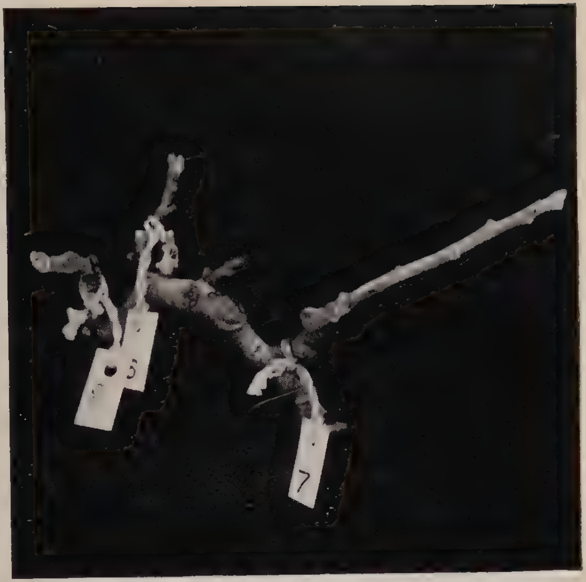

FIG. 19.

Spur B, November 7th, 1947. Note strong buds on spurs 7 and 8.

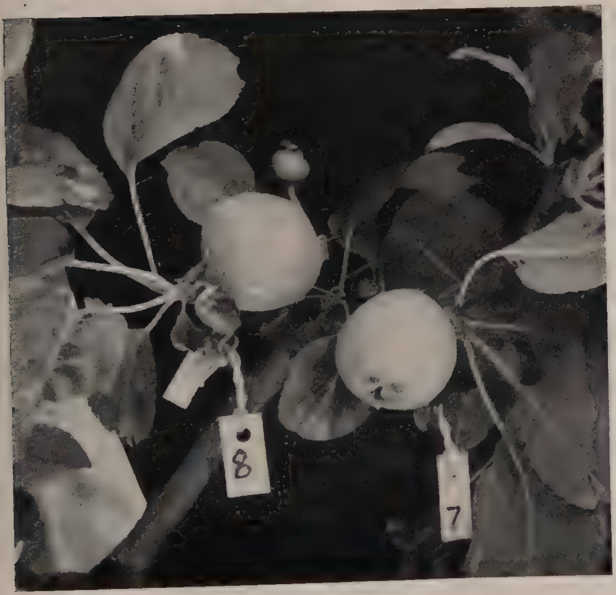

FIG. 20.

Spur B, July 16th, 1946. Note bourse bud clusters, and two primary leaves on spur 8, cf. Fig. 15.

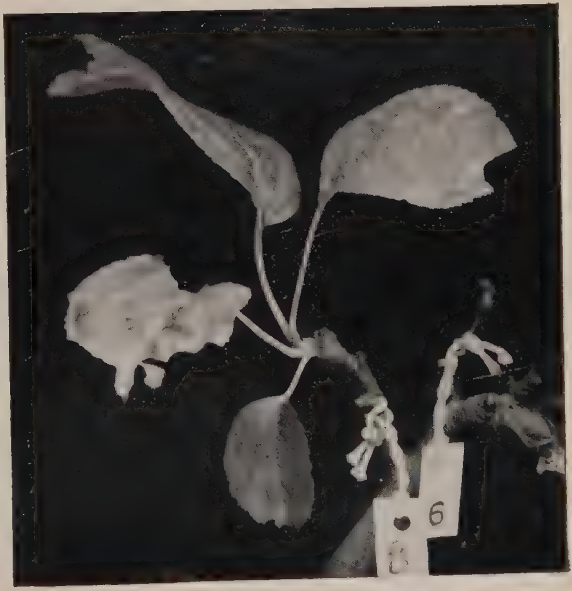

FIG. 21.

Spur B, September 27th, 1946. Note leaf scars left by shedding of primary leaves.

PLATE IV.

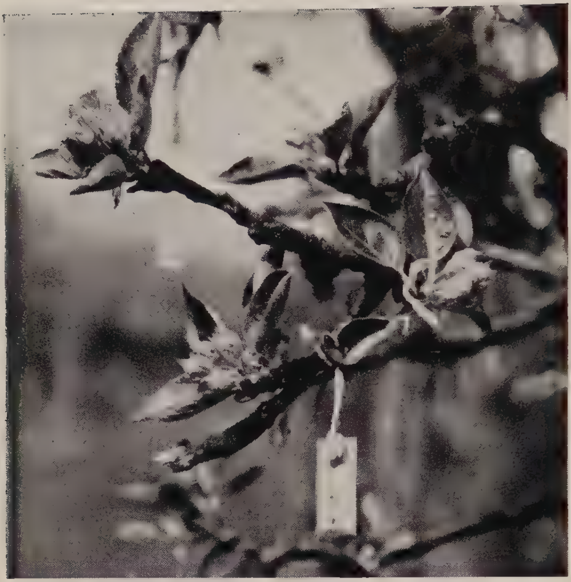

FIG. 22.

Spur C, *Ribston Pippin,* April 16th, 1946. Note blossoms and bold primary leaves.

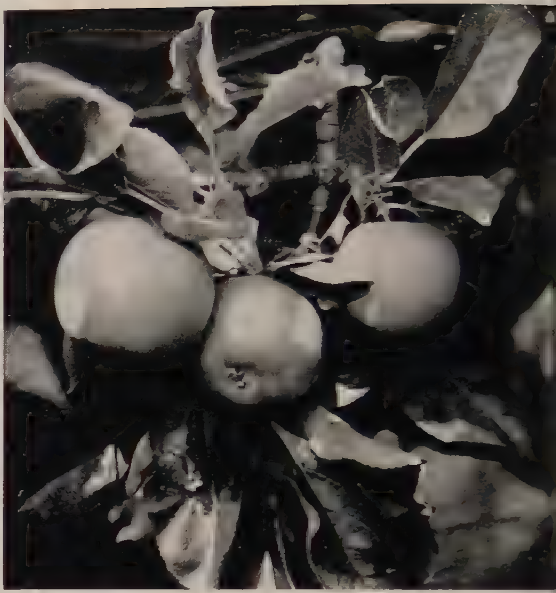

FIG. 23.

Spur C, August 26th, 1946. Note persisting prima
leaves and bourse buds at base of fruits.

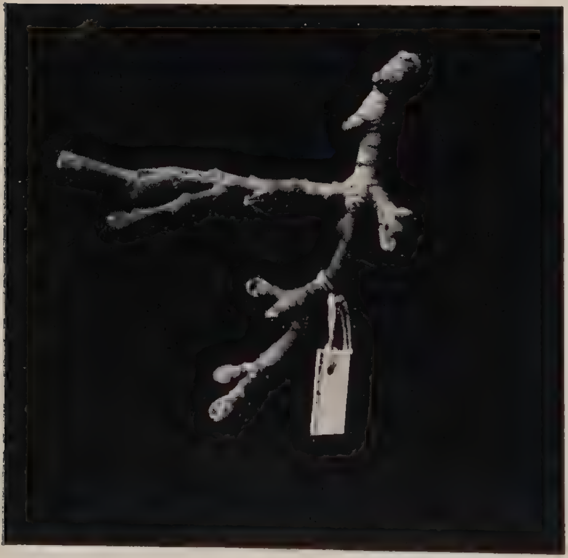

FIG. 24.

Spur C, April 2nd, 1947. Note bold bourse buds and short dard on left.

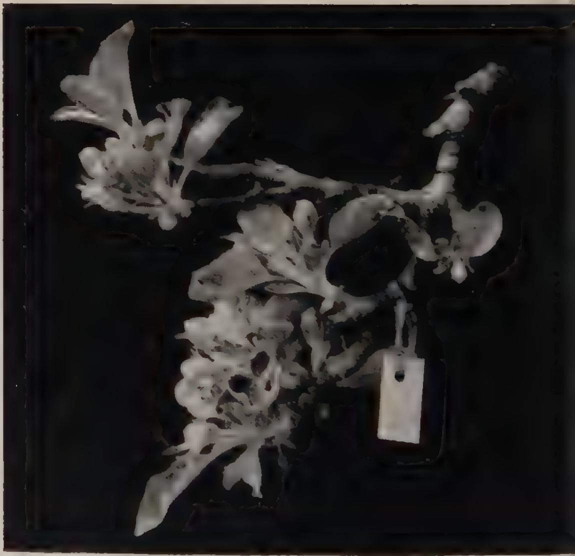

FIG. 25.

Spur C, May 2nd, 1947. Note blossoming, as in Fig.

PLATE V.

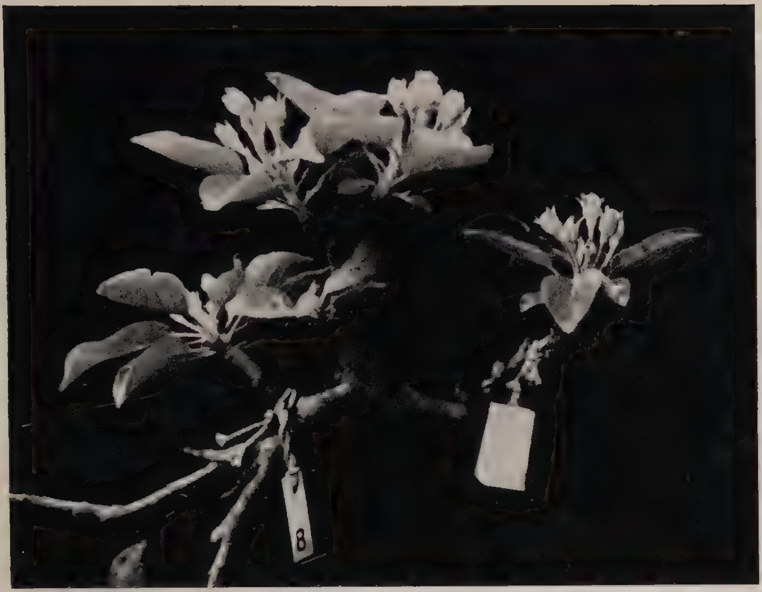

FIG. 26.
Spur D, *Ellison's Orange*, May 2nd, 1947. Note bold primary leaves.

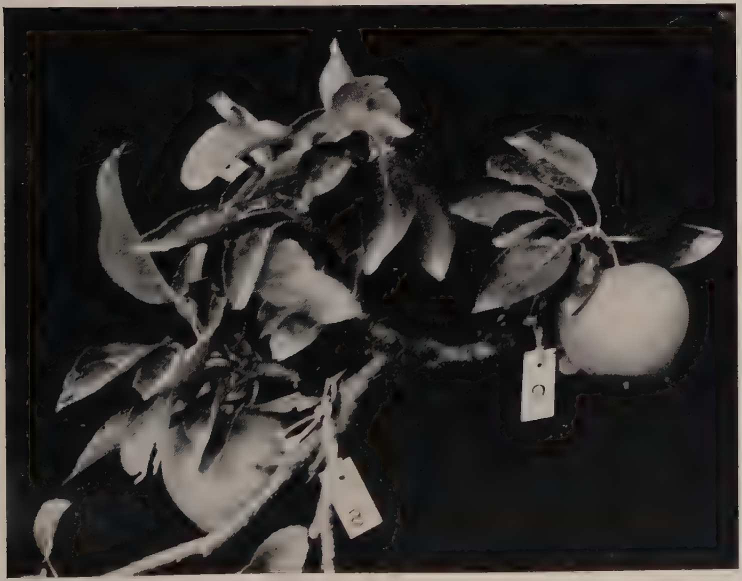

FIG. 27
Spur D, August 5th, 1947. Three large primary leaves and four bourse bud leaves can be seen on spur 9.

PLATE VI.

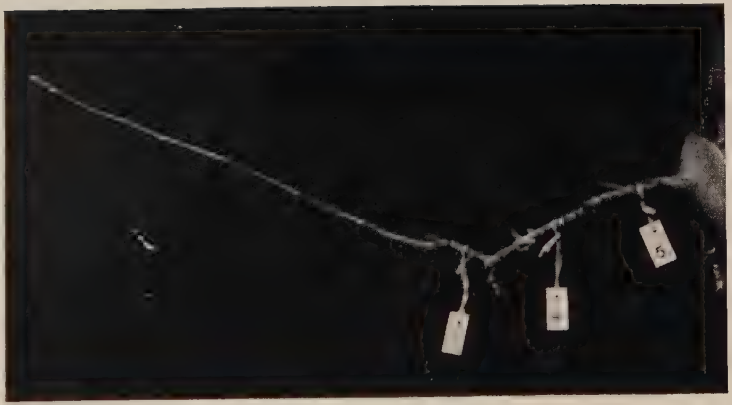

Fig. 28.

Blenheim Orange, November 25th, 1946. Note slender buds and shoot.

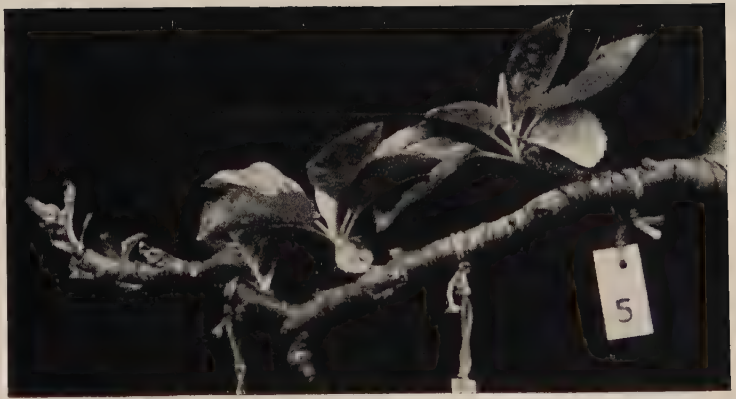

Fig. 29.

Blenheim Orange, May 2nd, 1947. Note only vegetative clusters.

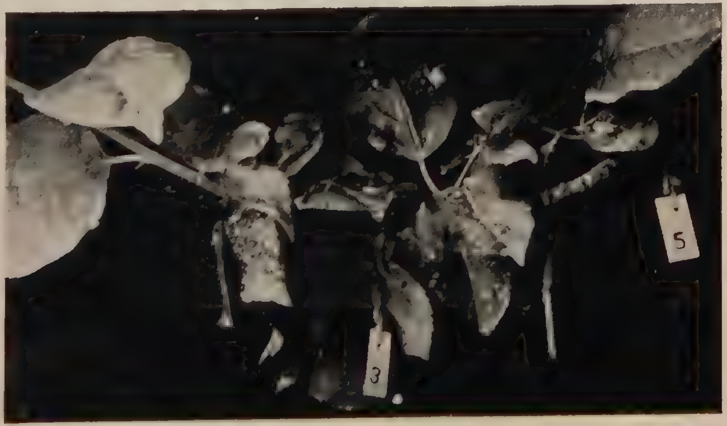

Fig. 30.

Blenheim Orange, June 10th, 1947. Note strong leaf production.

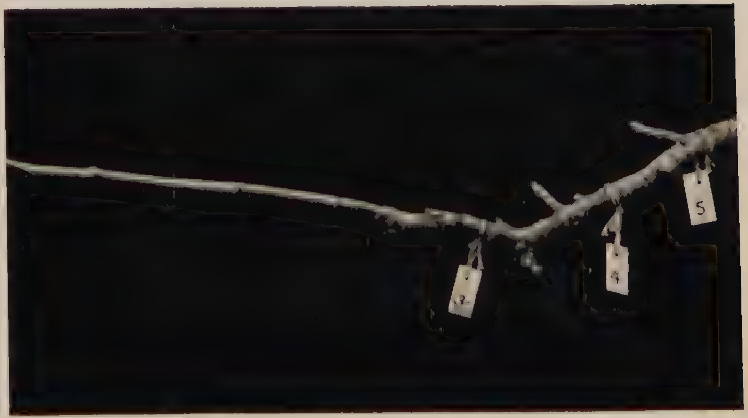

Fig. 31.

Blenheim Orange, November 7th, 1947. Note buds are again slender.

VERTICILLIUM WILT OF THE HOP *(HUMULUS LUPULUS)*

IV. STUDY OF A FLUCTUATING OUTBREAK

By W. G. KEYWORTH
East Malling Research Station, Kent

Fluctuating outbreaks of Verticillium Wilt of the Hop (caused by *Verticillium albo-atrum*), were first studied by Harris (1936) and later by Harris and Furneaux (1938), who showed that the intensity of symptoms as measured by leaf wilting and discoloration varied in different parts of the field and also from year to year. Symptoms were most noticeable in those parts of the field where underground springs induced local waterlogging and the disease tended to be more severe in wet seasons than in dry ones. Individual plants showing symptoms in one season might appear normal in the next year and vice versa. *V. albo-atrum* was isolated from plants not showing symptoms and it was concluded that the pathogen was widespread throughout the whole field but that symptoms appeared only in those areas where the soil was sufficiently moist during the summer months.

Keyworth (1942) distinguished another and more severe type of outbreak (" Progressive "), in which the disease spread from infection foci. Isolates of *V. albo-atrum* from outbreaks of the severe type were subsequently shown to be more virulent than those from fluctuating outbreaks (Isaac and Keyworth, 1948). These writers also noted that some fluctuating attacks caused milder symptoms than did others, the only sign of disease often being a few thickened bines (stems) on scattered plants.

The Ministry of Agriculture and Fisheries instituted in 1943 a scheme for the inspection of hop fields for freedom from Verticillium Wilt so that cuttings from such fields could be used as a source of planting stock free from the disease. The primary object of the scheme was to check the spread of the progressive type of Wilt, but owing to the difficulty of distinguishing between some fluctuating and progressive outbreaks by one inspection it was decided at first to exclude fields showing any symptoms of Wilt whatsoever from certification.

A study of a mild fluctuating outbreak was therefore made by the writer with the main object of determining where infected cuttings might be found in the field in relation to the distribution of visibly diseased plants. In order to follow up the work by Harris and Furneaux, observations were also made on annual variations in the distribution of visibly diseased plants and on the soil features of the field.

The field chosen for this survey was situated on the Weald plain, north of Brenchley, in Kent. It contained, in 1943, a number of plants showing mild symptoms of Wilt, mainly grouped in the eastern corner of the field (see text Fig. 1). Very few of the plants bore any wilted bines and in most cases the only external symptom was the thickened corky appearance of some of the bines. The wood in these bines was brown and usually the pathogen could be readily isolated from it.

1. DISTRIBUTION IN THE FIELD OF PLANTS WITH INFECTED CUTTINGS.

To determine the distribution of plants with infected cuttings in this field, sample plants were selected in the following categories :

(*a*) Showing symptoms. (Visibly diseased plants.)
(*b*) Adjacent to visibly diseased plants.
(*c*) In the same area but not adjacent to visibly diseased plants.
(*d*) In an apparently healthy area in the same field at some distance from the visibly diseased plants.

To provide for possible statistical analysis of the results, the plants in categories (*a*) and (*b*) were grouped in fours, each group containing two visibly diseased and two adjacent plants. The plants in category (*c*) were also in fours, but those in category (*d*) were selected at regular intervals over an area containing 1,880 plants (roughly $1\frac{1}{2}$ acres).

There were 32 plants in each of the categories (*a*), (*b*) and (*c*) and 64 in category (*d*). During the winter of 1943-44 all those cuttings which would normally be used for propagation (the basal, below-soil portions of the bines) were collected from each of the 160 test plants. It being desired to obtain an isolate from as near the base as possible, isolations were made from each cutting as follows :

All adhering soil was washed away and the basal $1\frac{1}{4}$ in. of the cutting removed. This portion was dipped in alcohol and flamed and the basal $\frac{1}{4}$ in. removed with a sterile scalpel. An incision was then made round the cutting to remove the next $\frac{1}{8}$ in. of cortical tissue. Two transverse sections were taken from the exposed wood cylinder and placed on potato-dextrose agar containing 1·0 per cent. potassium thiocyanate added to inhibit bacterial growth (McLean and Cook, 1941). It was necessary to take complete transverse sections as the fungus was often confined to a portion only of the vascular cylinder.

The plates were observed on several successive occasions during a period of 21 days and the presence or absence of *Verticillium* noted. In some cases it was difficult to be certain that *Verticillium* was absent owing to the more rapid growth of other fungi and also because the presence of potassium thiocyanate in the agar tended to suppress the production of *Verticillium* conidiophores, but previous tests of the method ensured a reasonably accurate diagnosis in most cases. Other fungi, which were frequently isolated and which were liable to be confused with *Verticillium* because of somewhat similar conidiophores were *Cephalosporium* sp. and *Gliocladium roseum*.

The isolation records are given in Table I.

TABLE I.

Record of isolations. 1943.

Plant category.	No. of plants.	No. containing *Verticillium*.	No. of cuttings.	No. containing *Verticillium*.
(*a*) Visibly diseased	32	29	216	95
(*b*) Next to (*a*)	32	11	230	24
(*c*) In the same area ..	32	5	231	6
(*d*) In a " healthy " area ..	64	2	456	2

This record indicates that the probability of obtaining infected cuttings is greatest from visibly diseased plants and that this probability decreases with increasing distance from such plants. A number of plants, however, in each of the three "apparently healthy" categories (*b*), (*c*) and (*d*) yielded infected cuttings and it is thus obvious that the distribution of the fungus was not limited to the plants showing symptoms. It is indeed probable that the fungus was more widely distributed than the results indicated and for this there are two reasons, viz. (*a*) the practical difficulty of guaranteeing a negative isolation record,* and (*b*) the fact that the fungus may have been present in the rootstocks even if not apparent in the cuttings.

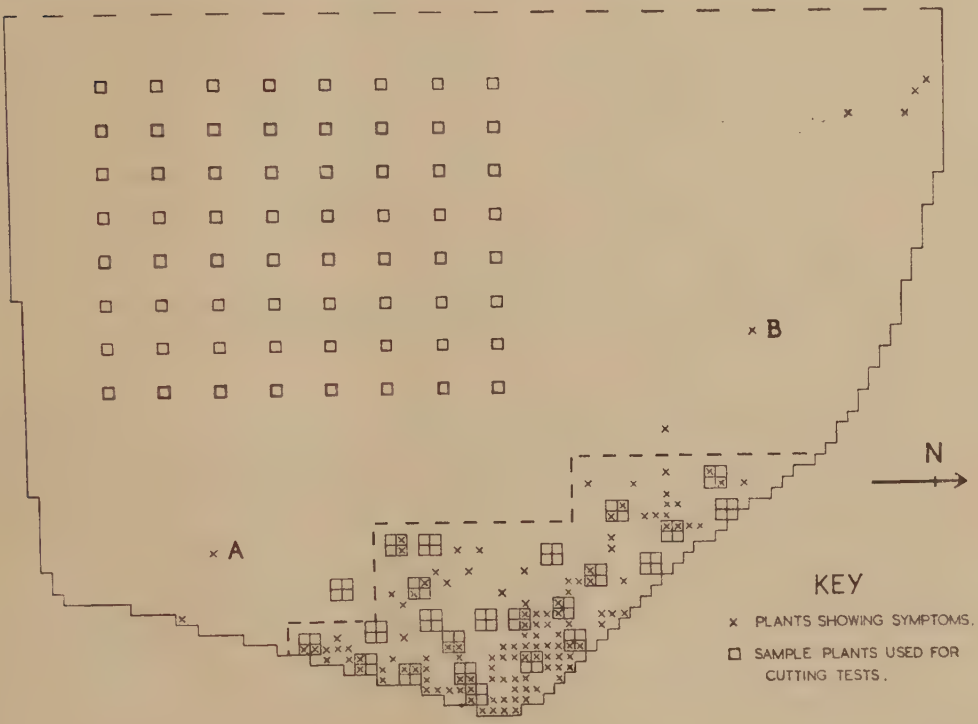
× B
× A
N
KEY
× PLANTS SHOWING SYMPTOMS.
☐ SAMPLE PLANTS USED FOR CUTTING TESTS.

FIG. 1.

Diagram of part of the hop-field studied, showing plants visibly diseased in 1943 and positions of plants used for isolation tests.

2. DISTRIBUTION OF PLANTS WITH INFECTED CUTTINGS AROUND ISOLATED VISIBLY DISEASED PLANTS.

Two visibly diseased plants were selected for this survey (marked A and B in text Fig. 1).

Early in 1944 the cuttings were collected from each of these plants and from the eight surrounding plants. In the case of plant B, cuttings were also taken from the next sixteen surrounding plants.

---

* The fact that no *Verticillium* was isolated from three visibly diseased plants may be taken as a measure of the fallibility of the method.

Isolation tests were made of all the cuttings by the methods described above. The number of infected cuttings from each plant and the disposition of these plants is shown in text Fig. 2.

These results indicate that although symptoms were visible on only the one plant in each locality the pathogen was present in the cuttings on several surrounding plants in both cases. No symptoms were seen in 1944 either on the two plants showing symptoms in 1943 or on the surrounding plants.

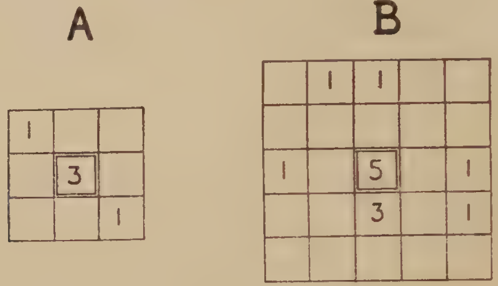

Fig. 2.

Diagram of positions of test plants around the two isolated visibly affected plants A and B. The numbers indicate the number of cuttings from each plant from which *Verticillium* was obtained.

3. INCIDENCE OF SYMPTOMS 1943-45.

As shown in text Fig. 1, the visibly diseased plants in 1943 were, with a few exceptions, confined to an area to the east of the field. The isolation record, however (p. 150 above), demonstrated that the pathogen was not confined to such plants.

The presence of *Verticillium* in the cuttings from apparently healthy plants might, of course, have indicated that such plants had only recently become infected and would possibly therefore show symptoms in 1944 or 1945, thus indicating a spread of disease. In fact no such increase in the number of plants showing symptoms was noted. Of the 93 test plants within the disease area (three of the original 96 having been removed in 1944), 31 appeared diseased in 1943, 27 in 1944, and 33 in 1945. Of all the plants in the field 130 showed disease in 1943 and 98 in 1944. Unless, therefore, there was a very considerable time-lag between infection and symptom expression, and experiments by Isaac and Keyworth (1948) show this to be unlikely, it may be concluded that the disease, as measured by the number of plants showing symptoms, was not spreading.

Although the number of visibly diseased plants remained approximately constant, the symptoms were not confined to the same plants each year and, within the disease area, the fluctuation in symptom expression on individual plants already described by Harris (1936) was noted.

The symptom history of the test plants in the three original groupings within the main disease area is given in Table II. The numbers in heavy type indicate either plants showing symptoms or those from which the pathogen was isolated and the numbers in light type indicate apparently healthy plants.

The total numbers of plants in each of the 1943 categories which showed symptoms in 1944 and 1945 are as follows. In category (*a*) 18 were visibly diseased in 1944 and 21 in 1945; in category (*b*) the numbers are 8 and 9; and in category (*c*) 2 and 4. It is thus apparent that although fluctuations in symptom

TABLE II.

Record of isolations and symptom history of test plants.

1943.		1944.	1945.
Category and No. of plants.	No. in which isolation was positive or negative.	No. in which symptoms present or absent.	No. in which symptoms present or absent.
(*a*) **31***	**28**	**16**	**12**
			4
		12	**6**
			6
	3	**2**	**2**
			0
		1	**1**
			0
(*b*) 30*	**10**	**5**	**3**
			2
		5	**2**
			3
	20	**3**	**0**
			3
		17	**4**
			13
(*c*) 32	**5**	**0**	**0**
			0
		5	**1**
			4
	27	**2**	**2**
			0
		25	**1**
			24

---

* Some of the original plants in these categories were removed in 1944.

expression occurred, these were not at random and there was a tendency for plants to retain their symptom status. It is noteworthy that the plants in categories (*b*) and (*c*) showing symptoms in 1944 and 1945 were not confined to those from which the fungus had previously been isolated.

The non-random nature of the annual distribution of visibly diseased plants is also demonstrated by a study of the total numbers of plants in the main disease area which showed symptoms in 1943 and 1944 and in both of these years. If, within this area, the presence or absence of symptoms on any particular plant were entirely a matter of chance, the proportion of plants showing symptoms in both years should approximate to the proportions showing symptoms in each year multiplied together.

Of the 508 plants in the area (see text Fig. 1), 122 showed symptoms in 1943, 87 in 1944 and 64 in both years. If such plants were distributed at random each year only 21 would have been expected to show symptoms in both years

$$\text{i.e.}\quad \frac{122}{508} \times \frac{87}{508} = \frac{21}{508}$$

and it is thus again obvious that there was a tendency for plants showing symptoms in one year to do so in the next.

4. Soil survey.

As the diseased plants were mainly aggregated in one corner of the field it seemed possible that soil conditions in this area might differ from those elsewhere. As has been stated, Harris (1936) noted that in the outbreaks he studied symptoms were more apparent in parts of the field where local water drainage impedance occurred. Evidence for this was obtained by a study of soil profiles, in which drainage impedance was indicated by soil mottling, and by observations on the height of water in perforated tubes sunk in the soil. It was not possible, in the present investigation, to obtain direct readings of water levels but a study of soil profiles was made in both the disease and " healthy " areas by means of 25 borings in each area made to a depth of 30 in. at regularly spaced intervals.

The field is level and the soil alluvial, the parent material being Hastings beds and Weald Clay. As is usual in such a soil, considerable variations in soil profile were apparent. The top 6-10 in. of soil were fairly uniform over the whole field, being a dark grey-brown silty loam containing a medium proportion of decomposed organic matter. There was then from 12-20 in. of light brown very fine sandy clay loam, passing at most sites into very fine sandy clay, both more or less heavily mottled and rust stained and containing a varying amount of iron oxide concretions and gravel. Layers of hard iron-stained dark brown sand and gravel were encountered at a depth of 24-30 in. at some sites in both areas. In twelve borings in the " healthy " area there was a seam of Weald clay material some 6-12 in. thick and generally at from 12-24 in. below the surface, the soil above this layer showing obvious signs of impeded drainage.

It was concluded that the soil over the whole field was badly drained, but no consistent difference in drainage or in any other respect detectable from soil profiles was noted between the two areas. It seemed likely that those parts of the field in which the seam of Weald clay occurred might suffer from local drainage impedance, but these sites were not in the disease area, nor were they representative of the whole of the " healthy " area.

DISCUSSION.

The annual symptom records demonstrate that the disease in this field (as measured by the numbers of plants showing symptoms) was not spreading. The record of isolations shows that the pathogen was present in the cuttings of many plants which appeared normal in 1943. It also seems likely that if it had been possible to test the rootstocks for the presence of *Verticillium*, an even wider distribution of the fungus would have been revealed.

The aggregation of most of the visibly diseased plants in the eastern corner of the field thus suggests that conditions in this area (presumably connected with the soil) were the main determinant of symptom expression, but confirmation of this could not be obtained by a study of soil profiles. Most of the visibly diseased plants noted in this area during 1944 and 1945 were among those showing symptoms in 1943, an intermediate number were among plants next to these and fewest among non-adjacent plants. This evidence suggests that there were environmental variations within the disease area and thus there was a tendency for a further grouping of visibly affected plants.

Subsequent work (Keyworth and Hitchcock, 1948) has shown that symptom expression on individual plants may be affected by the forward or late nature of the bine. Although all the bine in a hop field is theoretically the same in this respect, in practice variations do occur from plant to plant, more or less by chance. In addition to the main environmental factors already indicated there is thus a possibility that some chance effect such as this may influence symptom expression on individual plants.

SUMMARY.

The study of a fluctuating outbreak of Verticillium Wilt of the hop, during 1943-45, is described. The distribution of plants bearing infected cuttings was determined in relation to the distribution of plants showing symptoms in 1943 and the subsequent symptom history of these plants was noted. Some evidence that the pathogen was widespread throughout the field is given and it is concluded that the localization of the plants showing symptoms is due to environmental conditions. No evidence could be obtained of any correlation between this localization and soil structure.

ACKNOWLEDGMENTS.

The author is indebted to many colleagues who assisted in this work and particularly to Mr. F. C. Thompson of Wye College for advice on the soil features encountered, and to Mr. S. C. Pearce for his advice on the statistical interpretation of the results.

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REVIEWS

PLANT VIRUSES. By Kenneth M. Smith. 2nd edit. (London: Methuen & Co., Ltd. 1948. Pp. 78. Price 6s.)

Although described as a second edition of the book first published in 1935, this work has been completely rewritten and brought up to date and virtually nothing remains of the first edition. The general plan of presentation is similar to that used in larger works on the subject and the information is presented simply and clearly.

All of the main aspects of the subject are covered. There are chapters on the discovery and economic importance of plant viruses, the diseases which they cause, methods of transmission, insect vectors, the isolation and properties of the viruses themselves, serology, and the control of virus diseases. In the final chapter the various theories of the origin of viruses are summarized. In addition there is a useful bibliography of over a hundred titles.

The book is intended for those without previous knowledge of the subject and can be confidently recommended not only to students of plant pathology but to workers in other fields who may wish to have a short, authoritative and up-to-date outline of this rapidly developing subject. How rapid the development has been is brought out by even a brief consideration of the advances made since the appearance of the first edition. Methods of isolation, purification and crystallization of certain viruses have been evolved, and the electron microscope, with its new techniques, has been employed to throw fresh light on their physical properties. The increase in our knowledge of the physics and chemistry of viruses has been remarkable in such a relatively short time, but much progress has also been made in other fields, for example in the serology of viruses and in the study of the relationships of viruses to their insect vectors. It is all the more disappointing, therefore, to be reminded in reading Dr. Kenneth Smith's book that, in what might be called the " physiology " of viruses—their distribution in the plant, movement from the point of entry, multiplication, and effect on the plant's metabolism—we are apparently still where we were when the first edition appeared. And, while it cannot be said that the troubled waters of the problem of classification have been allowed to stagnate, little progress has been made in this direction either. In the first edition the author said, " Some system of classification of the large groups of plant viruses is an urgent need at the present moment " and in the present edition he says, " Some method of classification and nomenclature is urgently needed ". It has to be confessed that our knowledge of viruses remains largely empirical and it seems that we may have to wait for some time yet for a system which will " tie our facts into bundles ".

I.W.P.

GALL MIDGES OF ECONOMIC IMPORTANCE. Vol. III. FRUIT. By H. F. Barnes, M.A., Ph.D. Foreword by A. M. Massee, D.Sc. (London: Crosby Lockwood & Son, Ltd. 1948. Pp. 184. Price 15s.)

As Dr. Massee says in his Foreword to this volume, the literature dealing with the British Gall Midges, or Cecidomyiidae, was scattered through numerous and often inaccessible publications; the information concerning them was scanty and he might have added, but doesn't, that there were no very good collections of named specimens anywhere in this country and even the National Collection had scarcely any. Identifications were done from the galls or the type of damage caused by a species on a particular plant. In 1924 Dr. Barnes came down from Oxford and started worrying his friends about a suitable subject for research work ; induced to take up the neglected Gall Midges, he worked under Theobald and, in America, under Felt, and very soon was busy studying the biology and all the factors affecting it of a wide variety of species and also amassing a magnificent collection. Year after year we had a succession of reports detailing the results of his studies.

Now Dr. Barnes, so well qualified and equipped, is putting into more general circulation, with the aid of a courageous publisher, the more essential information condensed into a series of books of which this is the third ; it deals with the Gall Midges of Fruit Trees, 46 kinds in all, ranging from those grown in our own orchards to the more tropical Jak Fruit, Cherimoyer and Mangosteen. The total number of midges referred to seems to be about 130. Simplicity is the keynote and no very special knowledge is required to use the book ; the midges dealt with are arranged under the hosts attacked and to obtain possession of all the known facts about a midge—how to identify it, its life history, natural enemies and so on—little more is required than the ability to read. It seems as simple as that. The amount of information available varies with the insect (as is to be expected) and one naturally expects and finds it to be fullest where the author has been able to draw on his own studies. The correlation of the life history with the stage of development of the fruit attacked rather than with an average date, variable with district and season, is stressed so that control measures can be fitted into the normal routine. Distribution is adequate but will become more detailed with time ; the reviewer recalls the occasion when biological control was first in the air and how difficult it was to locate supplies of the Pear Leaf Midge (one of the first two insects to be investigated from this standpoint) in this country. Reading Dr. Barnes's account of this insect we see how much advance has been made since then.

It so happens that another pear species, the Pear Midge, illustrates a difficulty that so frequently confronts a student of almost any group of insects—the question of nomenclature. The author uses the scientific name of *Contarinia pyrivora* Riley, 1886, which is " almost certainly " the same as *nigra* Meigen, 1818 ; the latter would take priority over and would not be a synonym of *pyrivora* as the omission of a query mark has made it. We have been particularly interested in the boldness of the author, in his preface (p. 7), in proclaiming his faith about a " species " ; as both biologist and systematist Dr. Barnes is well qualified to express his opinion. To depart, however, from structure as a base for the separation of species sooner or later leads us into a morass of doubt and difficulty, and we feel that the biologist has not yet reached that stage of perfection in his technique that will induce us to place complete faith in his conclusions where they clash with pure morphology. The facts are given in their appropriate place in the book so that the grower is put on his guard. This is a book that simply must be in the library of every fruit-grower who wishes to keep abreast of his business and of every entomologist who aspires to something more than the narrow rut of specialization. L.

BOOKS RECEIVED

B.I.O.S. OVERALL REPORT No. 6. SOME AGRICULTURAL ASPECTS IN GERMANY DURING THE PERIOD 1939-1945. (H.M. Stationery Office, London, 1948, pp. 12. Price 4d. net.)

STUDIES IN BIENNIAL BEARING

IV. BUD-RUBBING, BLOSSOM-THINNING AND DEFOLIATION AS POSSIBLE CONTROL MEASURES

By L. B. SINGH
East Malling Research Station, Kent

THE seriousness of the biennial bearing habit in certain fruit trees, resulting in the alternation of a heavy crop in one year with little or no crop in the following year, has already been discussed in a review of the literature on the problem (Singh, 1948a), and some work designed to throw light on the causes of the habit was described in another paper (Singh, 1948b). In the present paper, the results of various treatments designed to break the biennial rhythm are described. These were carried out on trees of known biennial habit, some of which were in the " off " year phase and some in the " on ".

The main object of the treatments in the " off " year has been to lessen the formation of fruit buds and so prevent excessive cropping the following year. To this end, defoliation of the trees in different degrees and at varying times was carried out.

In the " on " year treatments the object was to encourage fruit-bud formation by reducing the crop at an early stage by such means as the rubbing out of fruit buds (called " bud-rubbing ") and by blossom-thinning. In addition an attempt was made to inject branches from both " on " and " off " year trees with the essential nutrients N, P and K to make good any deficiencies which may be connected with alternation of cropping. This gave negative results, but is briefly reported elsewhere (Singh, 1948c). The possible control measures tested may conveniently be dealt with in the following order :—

A. Defoliation in the " off " year.
  1. Branch defoliation at different times.
  2. Defoliation by chemical sprays.
B. Bud-rubbing and blossom-thinning in the " on " year.

A. DEFOLIATION IN THE " OFF " YEAR.

It seems reasonable to suppose that if the leaf area of vegetative spurs is reduced beyond a certain point, the carbohydrate supply may be restricted to a level at which fruit bud differentiation will be prevented. No conclusive evidence on these lines has yet been reported, although studies by Roberts (1923) and unpublished data obtained more recently by Mr. P. K. Sen at East Malling Research Station, suggested that this might be a useful line of approach to the problem.

There was no definite information available regarding the degree of defoliation necessary to ensure the maximum effect in checking fruit bud formation while at the same time maintaining the tree vigour. As it was feared that complete defoliation of branches might kill them, it was decided to practice only partial defoliation in the first place.

Similarly, as defoliation at different times would be likely to have different effects, a number of times of defoliation was tried, with the object of establishing which had the greatest effect upon the formation of fruit buds. The times of defoliation were chosen to cover the whole expected range of the critical period of fruit bud differentiation. The defoliation studies included three series of experiments, which will be discussed in order below.

(1) BRANCH DEFOLIATION AT DIFFERENT TIMES.

This study was performed in two experiments, (*a*) partial light defoliation at a series of times, on trees of Early Victoria, and (*b*) relatively severe defoliation at two periods on trees of Miller's Seedling.

(*a*) *Partial light defoliation on branches of Early Victoria.* Four trees of the variety Early Victoria (Syn. Emneth Early) in " off " year condition, were available for this experiment ; each of these trees had six to eight main branches forking from the trunk. In order to accommodate the six treatments which it was intended to test, with reasonable replication, each main branch was taken as an experimental unit. Six branches, of comparable girth at the base, were therefore selected on each tree and the treatments were distributed at random among these branches. In this way each treatment was repeated four times, once on each tree.

The treatments were untreated control and defoliation at five periods at intervals of 11 days, as follows : D1, 3rd May ; D2, 14th May ; D3, 25th May ; D4, 5th June ; D5, 16th June, 1946.

Method. On each occasion two out of every three fully expanded leaves were removed by hand or by means of scissors from every spur on the chosen branches. Since the majority of leaves had not fully expanded at the time of the first defoliation, it follows that the actual reduction in leaf area was slight on this occasion in comparison with that effected at the 3rd, 4th and 5th defoliations which were carried out at a time when all the leaves had fully expanded.

Records were taken to find the effects of the treatments on the vigour of the branches in the current year and their blossoming and fruiting performances in the following year. Measurements of branch vigour included records on (1) foliage development (increase in leaf number and area), (2) increase in girth, and (3) amount of new shoots produced. The records of blossoming and fruiting performances were on (4) the percentage of fruit buds as seen in spring, 1947, and (5) the number and weight of fruits produced. These are discussed in order below.

(1) *Foliage development.* A detailed record of the number of the leaves and their area was made at weekly intervals during the growing season, from 7th May onwards, on 20 selected comparable complex spurs of each treatment. The results are shown in Fig. 1. The dates when defoliation was actually performed are shown by the solid perpendicular lines. As the dates of leaf area measurement did not usually coincide with the dates of defoliation, the curves for leaf area of the samples have been projected from the nearest date of measurement, to give an estimated value for the leaf area immediately before and after defoliation. These parts of the curves are all shown with thin solid lines.

Both number and area of leaves follow the same general pattern, and show in the first place that all of the defoliation treatments considerably reduced the leaf area, for certain periods, and none of the defoliated branches subsequently reached the same maximum as the controls. The earlier the defoliation was carried out, the greater the degree of recovery in leaf development. There was relatively little recovery following defoliations in June but the recovery following the first defoliation (D1) was very rapid and striking; in fact three weeks later these branches could not be distinguished by eye from the control branches. This recovery was brought about by the unfolding of new leaves from the spurs. The second defoliation (D2) made 11 days after full bloom, appeared to result in a reduction of about half the leaf area from 15th May onwards, and this might be expected to have a greater effect than the other treatments.

(2) *Increase in girth.* The girth of the experimental branches, at 9 inches above the main crutch, was measured at monthly intervals throughout the season in 1946. A graph of the monthly increases, expressed as a percentage of the initial girth, showed that all the branches continued to grow at almost exactly the same rate irrespective of the defoliation treatment. This graph is therefore not reproduced here. The mean increase in girth of all the branches, by the end of the season, was about 4 per cent. and there was no significant difference between them. It was noticeable, however, that the branches defoliated 11 days after full bloom (D2) lagged behind the others from the end of July onwards. All the other branches actually slightly exceeded the control branches in their total percentage girth increase.

(3) *Amount of new shoot growth in 1946.* All the one-year-old shoots produced by the experimental branches were measured during the winter 1946-47. Later these shoots were pruned and a record was obtained of the pruning weights and of mean weight per shoot. In order to eliminate the effects of small initial differences in branch size, the data obtained were corrected to correspond with the mean branch girth. The corrections and the statistical analysis were done by analysis of co-variance on the logarithms of the original data and the corrected figures are given in Table I.

TABLE I.

Amount of shoot growth on branches receiving various defoliation treatments.

1946.	Wood growth* (cm. per branch).		Pruning weight* (gm. per branch).	
Branch treatments.	Total length (cm.)	Mean length per shoot (cm.)	Total weight (gm.)	Mean weight per cut shoot (gm.)
Defoliation D3 (May 25th)	4,457	29·85	957	5·74
Defoliation D1 (May 3rd)	4,667	34·91	938	6·84
Defoliation D2 (May 14th)	4,710	31·19	789	5·94
Defoliation D5 (June 16th)	5,309	35·56	912	6·27
Defoliation D4 (June 5th)	5,754	32·66	1,023	5·58
Control, untreated	6,012	36·39	1,197	7·21

The differences between bracketed figures are not significant.
* Corrected to correspond to a mean branch girth of 25·82 cm.

Table I shows that the branches receiving the first three defoliation treatments, D1, D2 and D3, produced a significantly smaller length of new shoots than the control branches. The reduction (corrected for branch girth differences) was of the order of 25 per cent. The later treatments D4 and D5 did not differ significantly from the control.

The differences in mean length of shoots, mean weight per pruning and total weight of prunings do not show any significant differences between the treatments, but it is interesting to note that treatment D2, which caused the greatest leaf reduction, produced the least weight of prunings, having a corrected weight about 34 per cent. below that of the control branches.

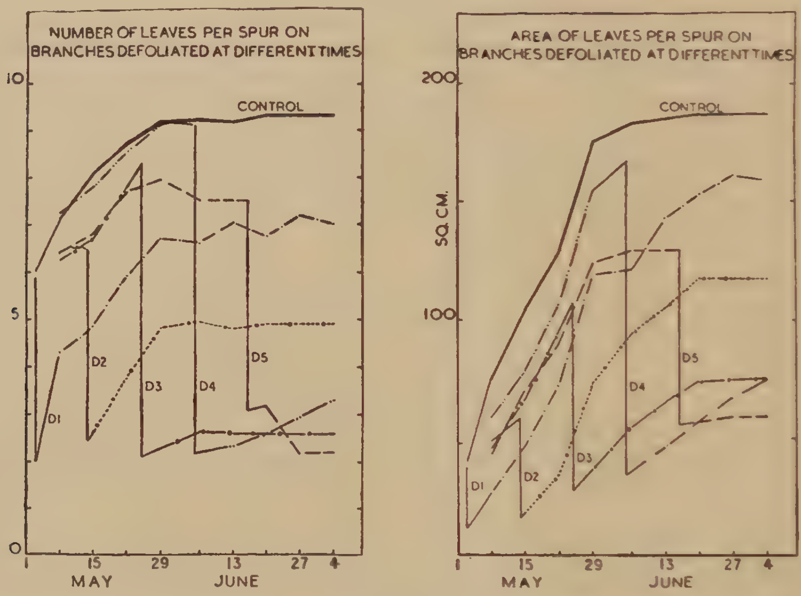

Fig. 1.

Number and area of leaves on spurs of Early Victoria, defoliated at different times.

(4) *Blossoming performance in 1947.* It is desirable to express the number of fruit buds in relation to the total number of buds present. This was done by counting both blossom and vegetative buds on the treated and control branches, about the middle of April 1947, when the fruit buds were just opening and the main leaf buds were only just beginning to break. The results are shown in Table II.

Table II indicates that a very high percentage of the buds on all the selected branches were fruit buds, and produced blossom in 1947. There was no significant difference in the percentage of fruit buds which, in fact, was over 94 per cent. in all cases. Thus it is seen that these branches of Early Victoria which had produced no blossom in 1946, all blossomed profusely in 1947, in spite of the leaf reductions shown in Fig. 1.

(5) *Fruiting performance in 1947.* The blossoms set well and produced a good crop. The fruits were thinned to singles. Picking was done on 30th July and 31st July and a record was taken of the number and weight of fruits per branch, from which

TABLE II.

Percentage of fruit buds and number and weight of fruits per branch (1947) following various defoliation treatments in 1946.

Branch treatments.	Fruit buds %	Number of Fruits.*	Weight of Fruit* (lb.)	Mean weight per fruit (lb.)
Defoliation D1 (May 3rd) . .	97·6	301	46·2	0·16
Defoliation D2 (May 14th) . .	96·2	353	56·6	0·15
Defoliation D3 (May 25th) . .	94·9	338	55·3	0·16
Defoliation D4 (June 5th) . .	99·1	304	49·3	0·16
Defoliation D5 (June 16th) . .	94·6	293	48·4	0·17
Control	97·5	335	57·3	0·17
Significance . .	N.S.	N.S.	N.S.	N.S.

* Corrected to correspond to a mean branch girth of 25·82 cm.

the mean weight per fruit was calculated. For statistical analysis purposes, the logarithms of the original data were taken and corrected for variation in initial size of branch, as in the case of wood growth. The corrected values are shown in Table II.

It can be seen in Table II that none of the treatments had any significant effect on number, weight or mean weight per fruit.

Conclusions. It is clear from this experiment that partial defoliation of individual branches of the variety Early Victoria had no effect in controlling excessive production of blossoms in the following season. There was also no effect on crop yield or fruit size.

The length of new shoots produced was reduced by defoliation during May, but branch thickness showed no significant difference.

It cannot be stated, from this experiment, whether one particular time of defoliation would be more effective than any other in modifying fruit bud formation, but the time of the defoliation which caused most striking reduction in leaf area was 11 days after full bloom.

(*b*) *Partial severe defoliation on branches of Miller's Seedling.* The main defoliation trial of 1946 was carried out on branches of "off" year trees of Miller's Seedling. In this experiment it was intended to test a more severe defoliation than that tried on Early Victoria and to have a better degree of replication. Thus the treatments were limited to three, i.e. two times of defoliation and untreated controls.

Four trees of approximately the same vigour were available for the purpose and, on each of these, six branches which were as comparable as possible, were selected for treatment. This gave a total of 24 experimental branches. As there were only three treatments, it was possible to have 8 replications of each and thus on each tree two branches received the earlier defoliation (D1), two the later defoliation (D2), and two were untreated controls. In nearly all cases there were also one or two extra branches, which were left untreated. The treatments were allotted at random so that each occurred once on northward- and once on southward-facing branches on each tree.

The times chosen for carrying out the defoliation were 7 and 22 days after full bloom (7th and 22nd May respectively). At these dates all the fully expanded leaves except one, on each spur and shoot, were removed by hand or with scissors, as described previously. Thus the degree of defoliation was more than twice as severe as that on Early Victoria.

Records were made on vigour and subsequent fruiting of each branch, as described for the previous experiment. These are discussed below:

(1) *Foliage development.* A fortnightly record of leaf number and area was taken on 20 comparable simple spurs from each treatment from the beginning of leaf expansion till autumn defoliation. The results are shown in Fig. 2, which is drawn in the same way as Fig. 1.

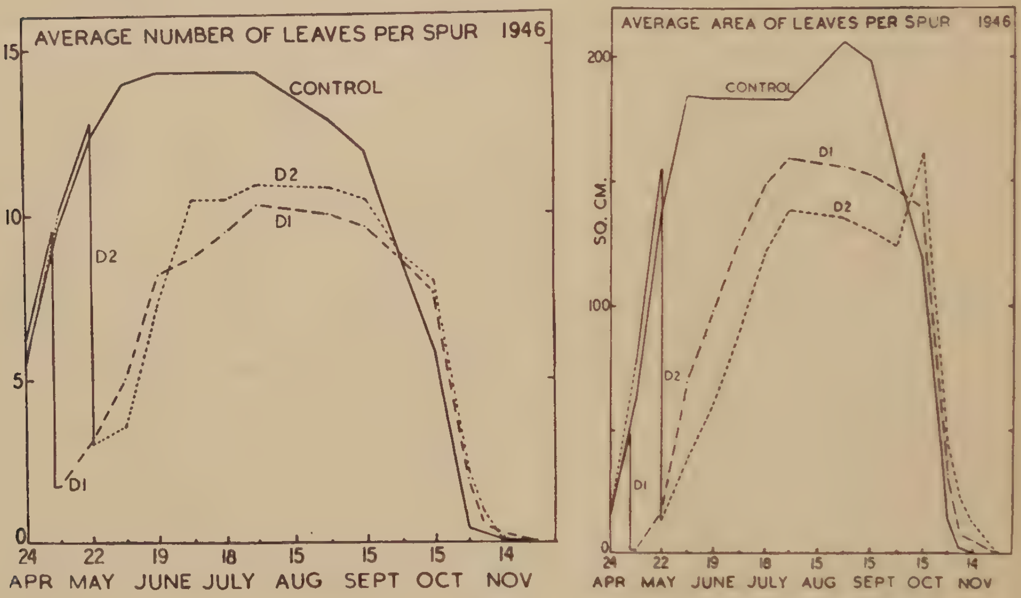

FIG. 2.

Number and area of leaves on spurs of Miller's Seedling, as affected by defoliation at two different times.

It will be seen that a spur before the first defoliation had an average leaf area of about 48 sq. cm. and this was reduced by defoliation to about 1 sq. cm. while those of the later defoliation had an average area of about 156 sq. cm. per spur which was reduced to 13 sq. cm. Although recovery was remarkably rapid, none of the defoliated spurs reached a leaf area equal to that of the control (undefoliated) ones during the growing season. Spurs on both types of defoliated branches reached their maximum area towards the end of July. At this time the D1 spurs had an average area of 159 sq. cm., the D2 spurs 128 sq. cm. and the control ones 183 sq. cm. The mean size of a fully expanded leaf at this time was 15·6 sq. cm in treatment D1, 12·7 sq. cm. in treatment D2 and 12·8 sq. cm. in the control.

The control spurs produced their maximum number of leaves by 19th June, and thereafter showed no increase of area until August when, probably on account of the wet weather, a little further leaf growth occurred. The leaves began to fall from the control spurs several weeks before those on the treated ones but, at the main autumn defoliation in October, the leaves on treated spurs were only a few days behind those on control ones.

(2) *Increase in girth.* The increase in girth of the treated and control branches was recorded at monthly intervals from 2nd April to November, 1946. Again, however, no striking differences in thickening of branches under the different treatments appeared. The total increase of the branch girth in both the defoliation treatments was slightly higher than that in the control but the differences were again not significant. However, it is clear that the defoliation treatment did not lessen the branch thickening.

It is of interest to note that in both Early Victoria and Miller's Seedling, the period of most rapid thickening at the base of the branches was from July to August. Thickening had ceased by the end of September.

(3) *New shoot growth in 1946.* Records of length of new shoots and weight of prunings were made, as in the Early Victoria trials. The values, corrected as before for initial differences of branch girth, are shown in Table III.

TABLE III.

Effect of branch defoliation on new shoot growth in the same season.

1946.	Wood growth.*		Pruning weight.*	
Branch treatments.	Total length (cm.)	Mean length per shoot (cm.)	Total weight (gm.)	Mean weight per shoot (gm.)
Defoliation D1 (May 7th) . .	769	18·2	147·9	3·0
Defoliation D2 (May 22nd) . .	286	15·4	74·3	3·0
Control	1,932	24·8	358·9	4·7

The differences between bracketed figures are not significant.

* Corrected to correspond to a mean branch girth of 23·71 cm.

Although very considerable differences between the treatments were found, unfortunately the variation from branch to branch, within a treatment, was so great that, even with the corrections, some quite large differences are not significant. Thus, although the total length of new shoots on the control branches was much more than twice that in either treatment D1 or D2, on branches of the same trees, this difference is not statistically significant. In the pruning weights, however, which also show a reduction of the same order in the shoot growth on the defoliated branches, the difference is significant and it is also significant in mean weight per cut shoot.

There is no significant difference between the two defoliation periods. It is clear, however, that defoliation at either period did materially check the new shoot growth.

(4) *Blossoming and fruiting performance.* A count of the fruit buds and the leaf buds produced on the branches was made at the pink-bud stage in the spring of 1947, in the same way as for the Early Victoria trees. Records were also obtained of the total number and weight of fruits carried by these branches and from these data the mean weight per fruit was calculated. The results are summarized in Table IV.

Table IV shows that defoliated branches produced about 15 per cent. less fruit buds than did the control ones, in the following spring. This reduction, however, was not statistically significant and the amount of blossom produced on the treated branches of Miller's Seedling was enough to enable the branches to set a heavy crop of fruits. There was again no difference between the effects of defoliation at the two periods.

TABLE IV.

Effect of branch defoliation on fruit yield in the following season.

Branch treatments. 1946.	Fruit buds %	Mean number Fruits.* 1947.	Mean weight fruits (lb.)	Mean weight per fruit (lb.)
Defoliation D1 (May 7th) ..	59·8	324	40·5	0·13
Defoliation D2 (May 22nd) ..	58·1	332	41·4	0·12
Control untreated ..	69·6	429	52·1	0·12

The differences between bracketed figures are not significant.

* Corrected to correspond to a mean branch girth of 23·71 cm.

The mean number and weight of fruits produced per branch are shown in Table IV. It is clear from this Table that the differences in fruit yield between the three treatments are of the same order as those in blossoming. The defoliated branches produced about 20 per cent. less fruits both in number and weight than the control ones but this difference was again not statistically significant. The mean size of fruits produced on the branches under different treatments was similar. In fruiting also, defoliation at the two periods respectively did not have any different effect.

DISCUSSION.

These two preliminary experiments in defoliation, using branches as the experimental unit, have given several clear indications of important effects which may follow leaf reduction at critical periods. Thus, leaf reduction in the early part of the season can reduce new shoot growth but apparently not branch thickening. Defoliation before about the end of May appears to cause more effective leaf reduction than does later defoliation. The effect of leaf reduction on fruit bud formation is less marked but it appears that defoliation of sufficient severity at a suitable time may tend to reduce fruit bud differentiation. None of the treatments tried in these two experiments caused a very large or significant reduction, however. The relative smallness of these effects may be due to one or more of several causes. In both cases treated and untreated branches were on a common root system, and any effect of the defoliation on the roots may have been partly or even entirely offset by effects from the untreated branches. Also, although it is known that in some ways the branches function independently, there may also be branch inter-action in other ways. Thus the branches which were not defoliated may have lessened the effects of defoliation carried out on other branches. The next step, therefore, was to continue defoliation tests, taking whole trees as the experimental unit. Such tests, on whole trees of Miller's Seedling, were started in 1947, and the results on the 1948 cropping and later performance of the trees will be available in due course.

(2) DEFOLIATION BY CHEMICAL SPRAYS.

Since defoliation of trees by hand, even if it succeeded in checking the formation of fruit buds in the " off " year, would be a long, laborious and expensive operation, it was decided to make a preliminary trial to find suitable chemical sprays which might bring about defoliation to the required degree without causing serious permanent damage to the trees. The writer has no knowledge of any previous work on defoliation of apple trees by means of chemical sprays, with the intention of preventing the differentiation of fruit buds. It was therefore decided to test various well-known sprays and other chemicals on the phytocidal effects of which a certain amount of information was available. Sprays of sodium chlorate, copper sulphate, tar oil and lime sulphur in various strengths, were tested and control trees were sprayed with water only.

Material and Method. The trees available formed originally part of a trial of new clonal rootstocks, bred at East Malling Research Station. They were budded in 1943 and thus were approximately three years old at the time of treatment. The varieties included were Cox's Orange Pippin and McIntosh Red.

The four different spray materials were each tested at four concentrations. Each spray was applied to four trees of comparable size, two of Cox's Orange Pippin and two of McIntosh Red. Control trees were sprayed with water. Thus 68 trees were included in the experiment.

The trees were sprayed on 10th May, 1946, at which time they all looked strong and healthy. All the trees were too young to blossom, but the leaves were green and were actively expanding. Young extension shoots had also started to grow.

The spray was applied with a small hand sprayer and a standard amount, sufficient to wet the leaves without excessive run-off, was applied to each tree. Neighbouring trees, by means of screens, were carefully sheltered from spray drift. The weather was bright and sunny but, on the following three days, became dull and windy with scattered showers.

The concentrations of the various sprays tested are shown in Table V together with details of observations on degree of scorch and defoliation in the subsequent month and on fruit bud formation as seen in the following spring.

(1) *Leaf scorching and defoliation.* Leaves of the sprayed trees began to show scorching two days after being sprayed. This differed according to the treatments and in some cases increased to the point of complete withering of the leaves. Most of the leaves of the trees sprayed with sodium chlorate withered up and died very quickly. The leaves on trees sprayed with copper sulphate showed typical small brown spots, while tar oil caused scorching of leaves in patches ; lime sulphur spray caused very little visible injury to the leaves. The trees of McIntosh suffered rather more damage than those of Cox.

A careful visual record of the scorching of the leaves was made on 20th May, ten days after spraying, each tree being classed according to the estimated percentage of leaves scorched. Both withered and scorched leaves remained on the tree for some weeks and then a number of them were gradually shed. Defoliation of the dead leaves in the sodium chlorate series started later than in the others and was relatively slow.

A visual estimate was made of the percentage of leaves that had fallen from each tree on 27th May, about five weeks after spraying, when it was considered that most of the leaves likely to fall off as a result of spraying (other than those remaining dead on the trees) had dropped. The results are set out in Table V.

Table V shows in the first place that sodium chlorate in all the concentrations tested, scorched nearly all of the leaves, while lime sulphur gave surprisingly little scorching. Even at 4 per cent., lime sulphur caused scorching of only 25 per cent. of the leaves. Copper sulphate and tar oil in the two higher concentrations scorched more than 50 per cent. of the leaves while the two lower concentrations caused less damage.

Similar results were obtained with regard to defoliation except that with sodium chlorate, the percentage defoliation was maximum (47·5 per cent.) with the lowest

TABLE V.

Results of a trial of various materials to cause scorching and defoliation of apple trees in order to reduce the number of fruit buds.

(1) Treatments.	(2) Spray.	(3) % strength.	(4) % leaves scorched (20.5.46).	(5) % leaves fallen (27.5.46)	(6) No. fruit buds per tree (May, '47).	(7) % fruit buds (May, '47).
A1 ..	Sodium chlorate	0·4	100·0	5·0	0·0	0·0
A2 ..	,,	0·2	100·0	17·5	0·0	0·0
A3 ..	,,	0·1	100·0	20·0	0·0	0·0
A4 ..	,,	0·05	95·0	47·5	0·0	0·0
B1 ..	Copper sulphate	0·8	80·0	17·5	7·0	4·0
B2 ..	,,	0·4	52·5	7·5	4·0	2·9
B3 ..	,,	0·2	35·0	2·5	1·5	1·4
B4 ..	,,	0·1	17·5	0·0	2·0	0·7
C1	Miscible Tar Oil winter wash	4·0	90·0	17·5	16·5	6·9
C2 ..	,,	2·0	47·5	5·0	0·0	0·0
C3 ..	,,	1·0	40·0	2·5	1·0	0·4
C4 ..	,,	0·5	12·5	2·5	1·0	0·3
D1 ..	Lime-sulphur (Sp. gr. 1·33)	4·0	25·0	5·0	16·0	17·6
D2 ..	,,	2·0	7·5	0·0	29·5	23·3
D3 ..	,,	1·0	2·5	0·0	33·5	19·0
D4 ..	,,	0·5	2·5	0·0	59·0	25·0
E ...	Water		0·0	0·0	46·0	25·7

concentration and minimum (5 per cent.) with the highest concentration. Presumably at the higher concentrations the leaves were killed so rapidly that they had no time to form an abscission layer.

(2) *Fruit bud formation.* Although it was hardly expected that the trees would form many fruit buds at only three years old, a count of the fruit and leaf buds on each tree was made in the spring of 1947. None of the McIntosh trees had any fruit buds, but a number were found on the Cox's Orange Pippin trees. Although the number of fruit buds is relatively small, they fall into a suggestive pattern and the data are therefore presented in Table V (columns 6 and 7).

It can be seen that, although 25 per cent. of the buds on the control trees were fruit buds, none of the buds on trees sprayed with sodium chlorate, at any of the concentrations tested, had differentiated into fruit buds. The trees sprayed with lime sulphur had percentages of fruit buds of the same order as the control trees, but the trees sprayed with copper sulphate had very much less.

Thus, although these data are not nearly sufficient to allow a final conclusion on the likely effect of these sprays on fruit bud differentiation, they nevertheless strongly suggest that certain sprays, in suitable concentrations may have a definite checking effect on fruit bud formation, and that, by choice of the material used, effects varying from nothing up to complete stopping of fruit bud formation may be obtained.

Further work on these lines, on mature trees, is clearly desirable, and should yield promising results.

B. BUD-RUBBING AND BLOSSOM-THINNING IN THE " ON " YEAR.

The artificial reduction of crop in an " on " year is one obvious line of attack on the problem of biennial bearing, and it has already been reported in paper II of this Series that, while fruit thinning has relatively little effect on inducing fruit bud formation, blossom-thinning has proved more promising (Singh, 1948a).

It has been suggested by Thompson (1941) that even earlier thinning of the potential blossom, by removal of fruit buds in the dormant period, may be a valuable method of converting irregular varieties to more regular cropping. A comparative trial, made at East Malling Research Station by Mr. N. H. Grubb who has kindly placed the unpublished data at the writer's disposal, showed that trees of Miller's Seedling which received spur shortening plus severe bud reduction in the winter 1940-41, preceding the " on " year phase, changed their phase of cropping and gave two successive " on " year crops, in comparison with trees receiving spur shortening only, the cropping of which remained relatively biennial.

It was therefore decided to test bud-rubbing and blossom-thinning of " on " year trees in such a way as to prevent a proportion of the spurs from fruiting in the current season, with the object of making them form fruit buds to crop in the normal " off " year and thus to break the biennial habit of the tree.

In the first experiment, in 1946, not enough " on " year trees were available to provide good replication of these treatments if applied to whole trees and it was therefore decided to use main branches of trees as experimental units as was done in the first defoliation experiments. In the following year, more material and more information were available, and therefore the treatments were applied to whole trees. The two bud-rubbing and blossom-thinning experiments were thus (1) on branches and (2) on whole trees. The latter experiment will be described later, when the results on fruiting are available.

(1) ON BRANCHES.

Material and Method. Four trees of Miller's Seedling apple, in Series A, in their " on " year in 1946 and of approximately equal vigour, were selected for the experiment. On each of these trees, six comparable branches were chosen ; thus twenty-four branches were made available for the three treatments, each of which had eight replications. Thus on each tree two branches received bud-rubbing, two blossom-thinning and two were untreated controls. The treatments were allotted at random as before, so that each occurred once on northward- and once on southward-facing branches on each tree.

The bud-rubbing of the branches was carried out on 28th March, 1946, at the bud-burst period when fruit buds could easily be distinguished from the leaf buds. This was 35 days before full bloom. The procedure adopted was to remove all the fruit buds from two out of each three successive complex spurs and all but one bud from the third, starting at the base of each branch and continuing upward to the top. The buds were broken off by hand.

For record purposes, the buds were weighed immediately in the field and later brought into the laboratory and counted. A count was also made of the fruit buds that were left on the branches. In all, 5,007 fruit buds weighing 750 gms. were removed from the eight experimental branches and only 808 fruit buds were left. Thus 86 per cent. of the total fruit buds were rubbed off. The treatment was rather drastic but was purposely made so in order to produce a definite effect.

Blossom-thinning was done 20 days later than the bud-rubbing, on 17th April, 1946, 15 days before full bloom. The blossoms which were in the pink-bud stage, were thinned to the same extent as in the bud-rubbing, i.e. all the blossom clusters were removed from two successive complex spurs and all clusters but one from the third. Here too, the clusters were pinched out by hand.

At the time of blossom-thinning, the primary leaves surrounding the blossom head had expanded, which gave this treatment a slight initial advantage of leaf surface over bud-rubbed spurs, in which the primary leaves were automatically removed along with the fruit buds. The total number of blossom clusters removed was 3,206 and the number left for cropping was 559, i.e. about 85 per cent. of the blossom clusters were removed. Thus about the same amount of blossom reduction was given by the bud-rubbing as by the blossom-thinning. Fruits on all the three treatments were thinned to singles in early June, as in the other experiments.

Detailed records on vegetative vigour and the subsequent fruiting performance of the branches were obtained and are discussed below.

(1) *Foliage development.* Forty comparable simple spurs on the bud-rubbed, forty on the blossom-thinned and thirty on the control branches were labelled and the number and area of leaves on them was recorded at fortnightly intervals. Half of the sample spurs on bud-rubbed and blossom-thinned branches were ones on which the blossom bud had been left. A record of their leaf number and area showed that they behaved practically like the fruiting spurs on the control branches, hence the foliage development of these spurs is not shown in Fig. 3 in which the leaf development per spur of the non-fruiting bud-rubbed and blossom-thinned spurs and the fruiting control spurs is presented.

It is clear from the graph that the bud-rubbed spurs had a smaller leaf area than the blossom-thinned and control ones until the first week in May, after which both the number and area of leaves was much greater on the bud-rubbed spurs than on the others. The blossom-thinned spurs had nearly the same number of leaves as the controls, all through the growing season, but their area was slightly greater from June onwards.

The increase in leaf area of the bud-rubbed spurs was due to the greater number of leaves upon them, which arose from numerous dormant buds which pushed out below the point where the fruit bud was rubbed out.

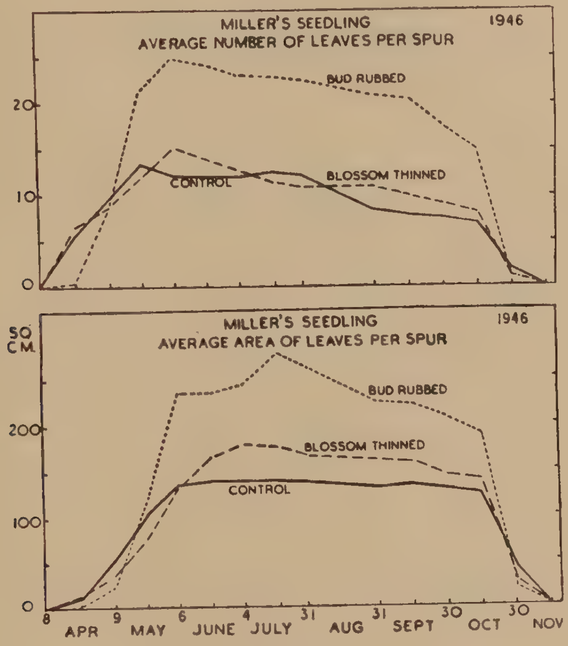

FIG. 3.

Number and area of leaves on control, bud-rubbed and blossom-thinned spurs.

(2) *New shoot growth.* The number and length of one-year-old shoots produced by the treated branches was recorded during the winter 1946-47 and a record was also obtained of the number and weight of prunings. The figures were corrected for initial differences in size of the branch, as in the branch defoliation experiment, and the corrected values for total length and mean length of shoot and similar data for prunings are presented in Table VI.

Table VI shows that there was no significant difference between the shoot growth of treated and control branches. The mean weight per pruning on the blossom-thinned branches was significantly less than on the bud-rubbed or control branches ; there seems no clear reason why this should be so and it may be a fortuitous result.

(3) *Increase in branch girth.* The girth of eight branches of each treatment was measured monthly, and their mean percentage increase, both in 1946 and 1947, is shown in Fig. 4.

It can be seen that in 1946, the year of treatment, the bud-rubbed branches thickened more rapidly than the others. In 1947, however, the control branches which were then in a completely " off " year condition, thickened more than the treated branches. Statistical analysis of both of these results showed no significant difference in the final increase of girth in each season, but the tendencies are interesting, especially in the comparison of bud-rubbed and blossom-thinned branches in 1946, which corresponds with differences in the leaf area.

(4) *Fruiting performance.* The number and weight of fruits from each branch were recorded and their values were corrected for differences in size of branch, as before. The corrected values for 1946, together with the mean weight per fruit, are shown in Table VII.

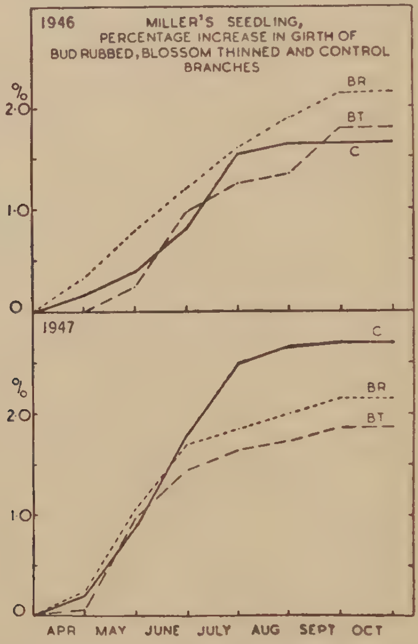

FIG. 4.

Branch thickening in 1946 and 1947 as affected by bud-rubbing and blossom-thinning in 1946.

It can be seen that both the bud-rubbing and blossom-thinning treatments greatly and significantly reduced the crop in 1946, bringing down the mean number of fruits per branch from over 400 to less than 65. This is not surprising, since over 80 per cent. of the fruit buds had been removed. The mean size of the fruits on treated branches was greater, as might be expected when the number was so much reduced. The bud-rubbing and blossom-thinning treatments in this year showed no appreciable differences in cropping.

The records of fruit buds and fruits, similarly corrected, on the same branches in the following year, 1947, are also shown in Table VII.

There was a large amount of blossom on both the bud-rubbed and blossom-thinned branches in 1947 while the control branches, as expected, had practically

TABLE VI.

Effect of bud-rubbing and blossom-thinning on new shoot growth in the same season.

1946.	Wood growth (cm.)*		Pruning weight (gm.)*	
Branch treatments.	Total length per branch.	Mean length per shoot.	Total weight per branch.	Mean weight per shoot.
Control	1,926	3·1	261·7	3·8
Bud-rubbed ..	1,584	2·8	260·7	4·4
Blossom-thinned	1,690	2·9	209·3	3·0

The differences between bracketed figures are not significant.

* Corrected to correspond to a mean branch girth of 25·47 cm.

TABLE VII.

Effect of bud-rubbing and blossom-thinning on branches of Miller's Seedling.

	Control.	Bud-rubbed.	Blossom-thinned.
1946 (Year of Treatment) :			
No. fruits per branch	425	64	62
Weight fruits per branch—lb. ..	**55·5**	**12·4**	**10·1**
Mean weight per fruit—lb. ..	0·13	0·17	0·19
1947 :			
No. fruit buds per branch ..	13	257	120
Percentage fruit buds	4·3	45·9	25·7
No. fruits per branch*	43	234	255
Weight fruits per branch—lb.* ..	**7·1**	**38·3**	**42·1**
Mean weight per fruit—lb.	0·15	0·17	0·17
Total weight 1946 + 1947	**62·6**	**50·7**	**52·2**

* Fruit figures are corrected to correspond with a mean branch girth of 25·47 cm.
Blossom figures are corrected to correspond with a total of 547 buds per branch.
The differences between bracketed figures are not significant.

none. The bud-rubbed branches, with 45 per cent. of their buds as fruit buds, significantly exceeded the blossom-thinned ones on which 25·7 per cent. of the buds were fruit buds. The control branches, with only 4·3 per cent. fruit buds, had significantly less than either treatment.

In number and weight of fruit also, the branches in both treatments significantly exceeded the control branches, producing about a bushel (40 lbs.) of fruit per branch, a normal good crop. The apples on the control branches, in spite of their low crop, were also smaller than those on the treated branches.

In total crop for the two years the control branches, with a total of 62·6 lbs. still slightly but insignificantly exceeded both the blossom-thinned branches, which had 52·2 lbs., and the bud-rubbed ones, which had 50·7 lbs. of fruit ; but since the fruit on the control branches was mainly of very poor size and quality, there is little doubt that the total market value of the fruit on the bud-rubbed and blossom-thinned branches greatly exceeded that of the fruit on the control branches, quite apart from the value of spreading a useful crop over two years.

(5) *Successive cropping of spurs.* A hundred fruiting spurs on each of the three treatments were labelled in 1946 with a view to finding whether any of them would crop regularly for two years in succession. A record of their cropping in 1947 showed that 3 per cent. of these spurs on control branches, 17 per cent. on blossom-thinned branches and 31 per cent. on the bud-rubbed branches cropped for a second year in succession. This is an interesting point, for it is unusual for Miller's Seedling spurs to crop in two succeeding years. The increase is very likely connected with improvement in general vigour of the branches, especially in relation to the leaf area. Of course, these spurs formed only a small proportion (of the order of 2 per cent.) of the total spurs which cropped in 1947. The rest of these were spurs which had been prevented, by the treatments, from cropping in 1946.

SERIAL PHOTOGRAPHS OF SPURS.

Serial photographs were made at monthly intervals in 1947, on typical blossom-thinned and bud-rubbed spur systems.

In Figs. 5-8 two spurs from which the blossom was removed, and one on which it was left, in the same spur system, can be seen. The untreated spurs gave typical " on " year performance and can be used for comparison with the treated ones. Another " control " spur from this experiment was illustrated in the previous paper (Singh 1948b, Figs. 10-18*) to which reference can also be made. In Fig. 5 the stage at which the blossom-thinning was done can be seen. The stalks of picked-off blossoms can be distinguished on the two deblossomed spurs. The other was left untreated. In Fig. 6, taken 20 days later, it can be seen that the primary leaves have persisted and have increased in size on the spurs from which the blossoms were removed, but they remained relatively small on the fruiting spur. Fig. 7 shows a strong foliage cluster which developed on the blossom-thinned spur, but the contrast in leaf area between deblossomed and fruiting spurs, as already discussed, is not so great as in the case of bud-rubbed spurs.

Finally, Fig. 8 shows the spur at the dormant stage, when the fruiting spur at the top has produced only a weak bourse bud, and the vegetative spur at the bottom, a well-developed bud. The third spur has remained very weak and therefore has not produced any prominent buds.

A bud-rubbed spur is seen in Figs. 9-12. In Fig. 9*a*, taken immediately after the operation on 1st April at the period of bud burst, the scars left where the buds were removed can be seen (compare Singh, 1948b, Fig. 10). Fig. 9 b, taken 30 days later, shows that adventitious buds had just started to grow from below the points where the original buds were rubbed out. Comparison with Fig. 5, taken the same day, shows that the control and blossom-thinned spurs had a much greater leaf area at this stage. Figs. 6 and 10 show the spurs twenty days later, at the time of petal-fall.

* By error, the blocks of Figs. 16 and 17 were transposed in the paper as it appeared in this Journal (Vol. 24, Plate II following p. 148).

It can be seen that during these twenty days the leaves on the bud-rubbed spur have developed rapidly and already exceed those on the fruiting one in size and number.

Fig. 11 shows the abundant foliage produced by August. (A comparable fruiting spur on the same date is shown in Singh, 1948b, Fig. 16*.)

A comparison at the end of the season is seen in Figs. 8 and 12. The weak bud formed on the fruiting spur (Fig. 8, top) can be compared with the bold fat buds on the bud-rubbed spur (Fig. 12). These in all probability are fruit buds, as the work described on page 172 has shown. One bud was left to blossom on one out of each three spur systems, both in the bud-rubbing and blossom-thinning trials, as already described. In each case, this bud gave typical " on " year performance, as shown here in spur G. (Fig. 8, top).

Thus these two series of photographs show clearly that, on spurs bearing fruits, the leaf development is relatively restricted but if fruit is prevented from forming, by removal either of blossoms or of the whole bud, a greater development of leaf generally occurs and this is responsible for the laying down of new fruit buds.

DISCUSSION.

Partial defoliation of certain branches of trees in the " off " year reduced the amount of shoot growth but did not reduce the branch thickening or fruit bud formation. Defoliation of a rather more severe nature had a similar effect on shoot growth and branch thickening but gave a reduction of the order of 15 per cent. in fruit bud formation, which was not significant.

On the other hand, complete defoliation of young trees carried out by sodium chlorate, prevented fruit bud differentiation while partial defoliation by tar oil and copper sulphate also checked it.

Thus a certain amount of defoliation appears to have little effect on fruit bud differentiation but beyond this amount it begins to lessen it. If, in addition, the growing tips are removed or injured, as in the case of spray treatments, the effect becomes greater. This suggests that the young developing leaves left on the partially defoliated trees may play an important part in providing the materials which cause fruit bud differentiation, and this in turn suggests that hormones may be, in part, responsible. No final conclusion can be drawn from the present experiment but further work on these lines should be well worth while.

With regard to treatments in the " on " year, it is already known that complete deblossoming of trees can change their phase of cropping, and so it would be possible to have some whole trees in " on " and some in " off " year conditions each year. This would not be satisfactory, however, since the heavy crops borne by biennial bearers in their " on " years would still be of poor quality.

The bud-rubbing and blossom-thinning treatments tested have proved much more successful, however, for they have produced a useful crop of good quality fruits on the same branches for two years in succession with the promise of a further satisfactory crop to follow. Looking back on the results, it appears that equally good or even better effects might have been obtained with a rather less drastic reduction in buds or blossoms in the year of the treatment.

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[*] By error, the blocks of Figs. 16 and 17 were transposed in the paper as it appeared in this Journal (Vol. 24, Plate II following p. 148).

Bud-rubbing, by hand, is more practicable than blossom-thinning by hand, and, especially if a less severe bud-thinning is sufficient, may quite readily be performed at the time of pruning. It is an advantage, however, to choose a time when the fruit buds can readily be distinguished from leaf buds.

For blossom-thinning, and possibly indeed for bud reduction also, it may yet be possible to find sprays which will reduce the hand labour required without causing damage to the trees.

In conclusion, this work has shown that the local conditions of each spur can provide limiting factors which govern its fruit bud formation. The number and area of leaves, in particular, appear to determine whether a spur differentiates a fruit bud or not. It appears that the desirable balance between growth and cropping can be established by regulating the local conditions of spurs by treatments such as bud-rubbing and blossom-thinning and that this will result in the production of a satisfactory crop in what would have been an " off " year, as well as a reasonable crop in the year of the treatment.

Long term experiments will be required to establish the duration of the effects of such treatments and to find the most satisfactory degrees of treatment.

SUMMARY.

1. Partial defoliation of branches of Early Victoria, at certain periods, reduced the new shoot growth but had no significant effect on either trunk thickening or fruit bud formation.

2. Rather more severe defoliation of branches of Miller's Seedling had similar effects on shoot and trunk thickening. A small reduction in fruit bud formation was not significant.

3. A preliminary trial of defoliation by chemical sprays, using sodium chlorate, copper sulphate, tar oil and lime sulphur, at various concentrations, showed that all degrees of leaf destruction up to 100 per cent. can be obtained. Certain sprays also checked fruit bud formation. Sodium chlorate, even at 0·05 per cent. scorched almost all the leaves and entirely prevented fruit bud formation. Lime sulphur had practically no effect. Tar oil and copper sulphate were intermediate. Further work on these lines appears promising, but it would be premature to make definite recommendations.

4. Bud-rubbing and blossom-thinning, resulting in removal of about 80 per cent. of the blossoms, on sample branches, improved their vegetative vigour, particularly in relation to leaf area. Bud-rubbing was more effective than blossom-thinning in this respect.

5. The crop on the bud-rubbed and blossom-thinned branches was much reduced in the year of treatment but in the following year the treated branches produced a normal crop of about a bushel (40 lbs.) per branch, in what would normally have been an " off " year.

6. It is suggested that by the use of such treatments, or modifications of them, a proper balance between growth and cropping can be established and satisfactory crops of good quality fruit maintained from year to year.

Studies in Biennial Bearing. IV. Bud-rubbing, Blossom-thinning and Defoliation as Possible Control Measures. By L. B. Singh.

PLATE I.

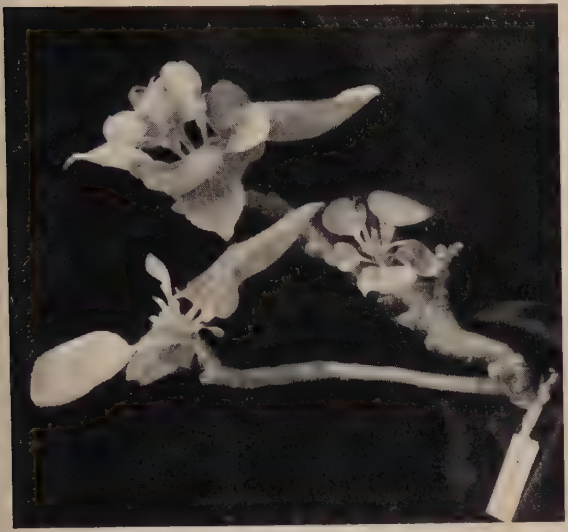

Fig. 5.

Miller's Seedling, Spur G, 1st May, 1947. Two blossom-thinned spurs and one untreated (at top). Note stalks of picked-off blossoms.

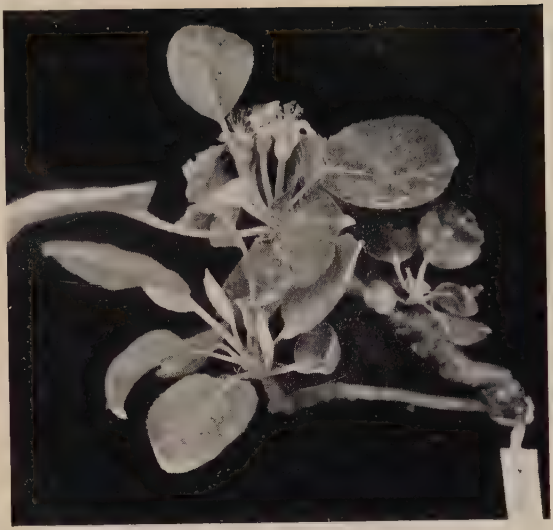

Fig. 6.

Spur G, 21st May, 1947. Note fruitlets on untreated spur, and new leaf development on one treated spur. The other remains weak.

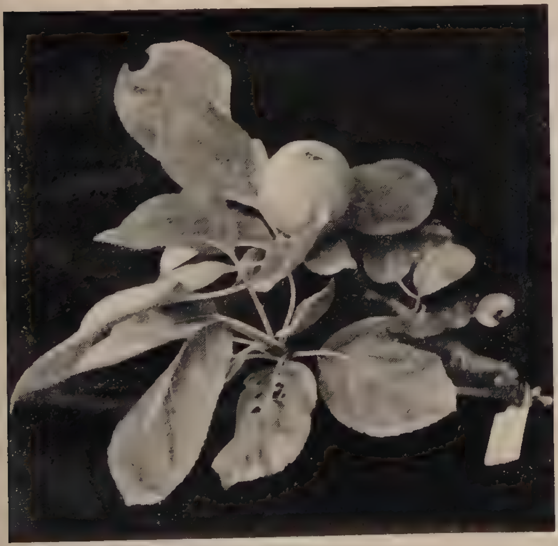

Fig. 7.

Spur G, 3rd July, 1947. Note strong vegetative cluster on treated spur.

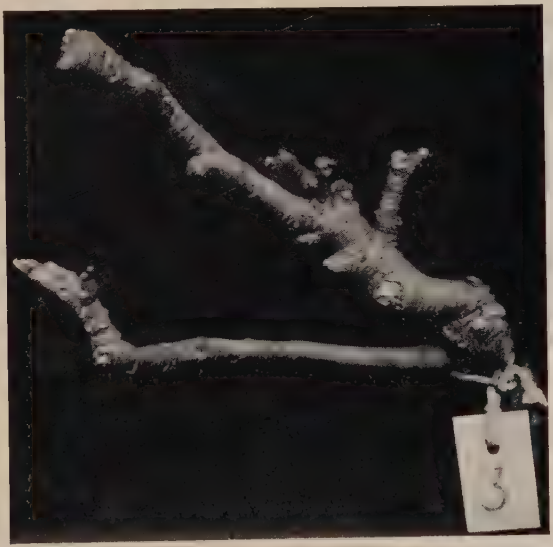

Fig. 8.

Spur G, 7th November, 1947. Note weak bourse-bud on fruiting spur and plump, well-developed bud on vegetative spur.

[*face p.* 176

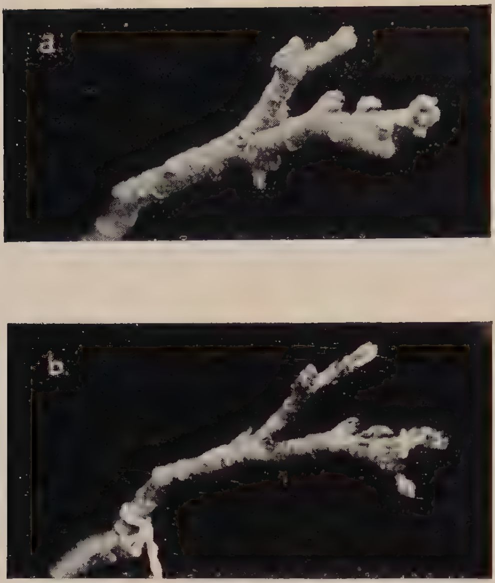

FIG. 9.

Miller's Seedling, Spur H. (*a*) Bud-rubbed, 1st April, 1947. Note bud scars. (*b*) 1st May, dormant buds beginning to grow.

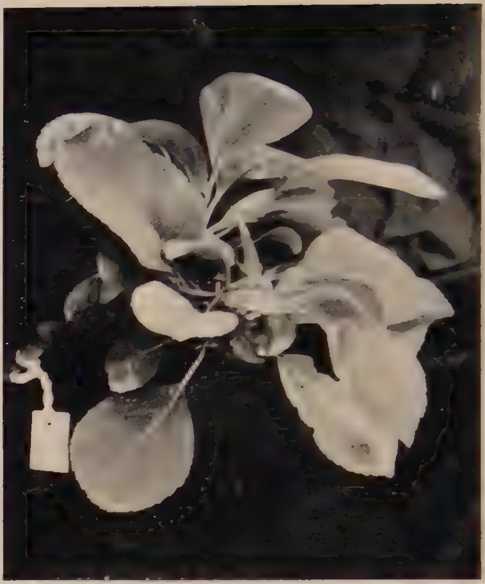

FIG. 10.

Spur H, 21st May, 1947. Note rapid growth of new leaves. Compare Fig. 9.

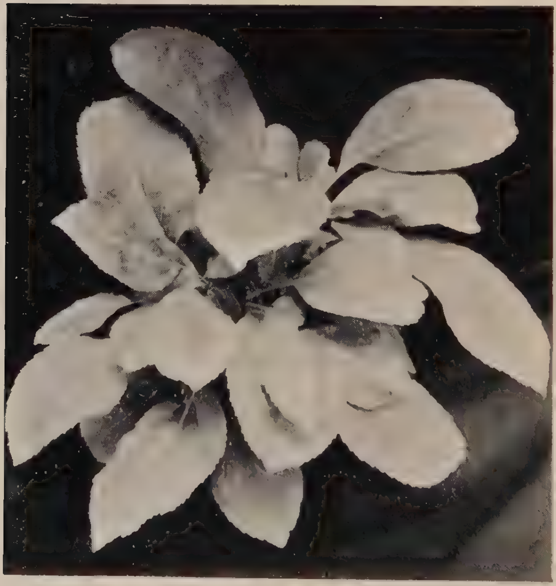

FIG. 11.

Spur H, 5th August. Note abundance of well developed leaves

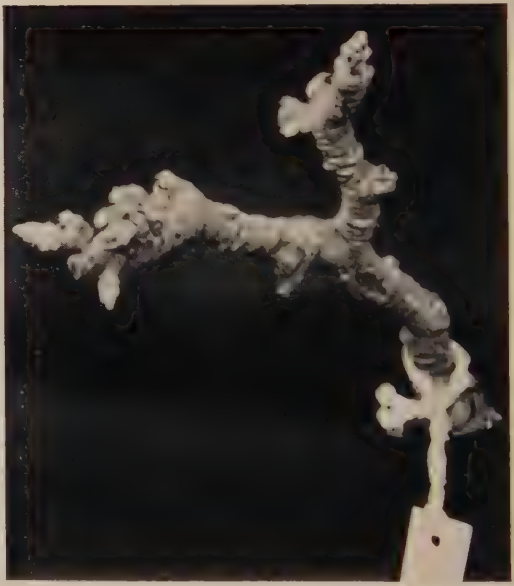

FIG. 12.

Spur H (from opposite side), 7th November, 1947. Note well-developed buds. Compare Fig. 8.

ACKNOWLEDGMENTS.

The author expresses his grateful thanks to Dr. R. G. Hatton, Director of East Malling Research Station, for providing the experimental material and facilities and to Dr. W. S. Rogers who supervised these trials, for his constant help and advice. Thanks are also due to Dr. M. C. Vyvyan for advice, to Miss F. Bacon for help with records and diagrams, and to Miss M. H. E. Davies for correcting the proofs.

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(*Received* 17/3/48.)

STUDIES OF THE PHYSIOLOGICALLY-ACTIVE VOLATILE ORGANIC COMPOUNDS PRODUCED BY FRUITS

I. THE CONCENTRATIONS OF VOLATILE ORGANIC COMPOUNDS OCCURRING IN GAS STORES CONTAINING APPLES

By **JOHN C. FIDLER**

Department of Scientific and Industrial Research (Food Investigation), Ditton Laboratory

SYNOPSIS.

When apples begin to ripen after gathering, volatile products are given off. Some of these account for the smell of the fruit ; another is the gas ethylene. These volatile products affect the rate at which the apples ripen, and are probably also responsible for such diseases of storage as superficial scald and spotting of the lenticels. The volatile products tend to collect in the atmosphere of the store and hence also in the fruit, particularly with tight packing or when the ventilation is restricted, as in gas storage. It is necessary to use oiled paper wraps in gas-storage, to prevent scald, but this is a laborious and somewhat costly procedure. It is hoped to reduce the amount of the volatiles in the atmosphere of the store, and thus to control scald, by employing a filter containing some material to absorb the volatiles. The filter would have the atmosphere of the store circulated through it, and could be fitted either inside or outside the store.

This paper describes an investigation, the first of its kind, of the amounts of the volatile products occurring in the atmosphere of gas-stores under present methods of operation. The main feature is the suprisingly high concentration of ethylene found in the stores. The amount present would, in ordinary air, be expected to cause stimulation and acceleration of ripening.

This raises the question whether, if ethylene could be removed, further benefits in delay of ripening could be obtained.

Introduction and Historical Survey.

Fruits, especially when ripe, give off a number of volatile organic substances. The rate of evolution of these substances (which in this series will be termed the " volatiles ") is usually very low. Certain of the volatiles are compounds such as esters or terpenes, responsible for the characteristic odours and aromas of ripe fruits. Their nature was first investigated, using apples, by Power and Chesnut (1920), who identified the methyl, ethyl and amyl esters of formic, acetic, caproic and caprylic acids. Later (1922) they identified geranial in ripe apples, and linalyl esters in ripe peaches. Thomas (1925) found acetaldehyde and ethyl alcohol in the apple volatiles.

It has been known for a long time that ethylene hastens the ripening of unripe fruits, and that some ripe fruits produce a gas which has the same effect. Gane (1934) identified this physiologically active volatile as ethylene.

The acceleration of ripening by ethylene is not the only form of physiological activity induced by the volatiles. Kidd and West (1938 b) showed that the spotting of the lenticels of late-maturing apple varieties, stored in the same chamber as early-maturing varieties, was caused by a high concentration of ethylene. This was confirmed by Smock (1942). Brooks *et al.* (1928) have suggested that apple scald is caused by the accumulation, near the skin of the apple, of one or more of these volatiles. This theory has been widely accepted, and the good measure of control of apple scald obtained by the use of the familiar oiled wrapper is generally held to be due to absorption of the physiologically active volatiles by the oil.

In some seasons scald can be very serious on English apples from certain localities, and its severity is aggravated by gas storage unless preventive measures are taken. While wrapping in oiled paper (if done at the right stage of ontogeny) is effective, it is not an ideal solution of the problem. Not only is it laborious and costly, but its great demerit is the amount of time it takes. For gas storage the fruit should always be got into store as quickly as possible after picking.

That the oiled wrapper was not the ideal method of control was always realized, and experiments to find new methods were begun by the Department of Scientific and Industrial Research before the war. These tests included the use of oil sprays to eliminate the volatiles ; they were, however, inconclusive and, together with the survey work begun by Walls (1942), had to be abandoned for more urgent war-time problems.

Gane's work had, however, stimulated workers in other countries. Thus Foulon (1944) tells how Fontanel (1936) was led to use a filter of active carbon in an attempt to remove ethylene from commercial cold stores containing pears. Fontanel did not succeed in this attempt, since activated carbon is not an ethylene absorbent at ordinary temperatures (see Walls, l.c., p. 64), but he did produce an efficient apparatus for deodorising cold stores; Fontanel's method is now a standard American practice. Smock (1943, 1944) and Southwick and Smock (1943) have also used active carbon for removal of volatiles, and report good elimination of ethylene and control of scald if the carbon is used as a carrier for bromine.

Several workers have made estimations of the rate of production of volatiles by fruits. Kidd and West (1938a) appear to have been the first, estimating total volatiles as carbon by passing the gas stream from the fruit through a combustion tube, and absorbing the carbon dioxide produced, the carbon dioxide of respiration being first removed by absorption in soda lime. Walls (1942) has objected that soda lime will absorb volatiles, but Potter and Griffiths (1946) state that soda lime does not absorb ethylene and quickly becomes saturated by the odorous volatiles, after which it does not absorb them. Since the amount of the odorous volatiles absorbed probably depends to some extent on their concentration, on temperature and possibly on relative humidity, the uncritical use of soda lime must introduce an unpredictable error, but it is likely that results obtained by the combustion tube method give fairly good indications of the rate of production of the volatiles.

Hansen and Christensen (1939), Christensen, Hansen and Cheldelin (1939), and Nelson (1940) all describe methods for estimating the ethylene content of tissue, and the rate of production of ethylene. Their methods are reliable and selective, but are not suitable for application to the continuous estimation of ethylene in an air stream.

Gerhardt and Ezell (1939) trapped the volatiles by absorption in sulphuric acid. Southwick (1945) has used a similar method. These workers determined the volatiles in the traps by oxidation with ceric sulphate, expressing the results either as equivalents to the amount of acetaldehyde oxidized by the ceric sulphate in a standardization run, or in terms of " mg. of ceric sulphate reduced ". The reason for this is that ceric sulphate does not oxidize organic compounds stoichiometrically, although the method can be made stoichiometric for ethylene.

Walls' (1942) method is a decided advance on this, in that the volatiles may be given as " mg. carbon ". He states that pure sulphuric acid absorbs the odorous fractions, and that sulphuric acid containing silver sulphate absorbs ethylene. These traps are oxidized by the method of Birkinshaw and Raistrick (1931), a wet oxidation with chromic acid. Walls found, using apples in air at about 15° C., that ethylene contributed about two-thirds of the total carbon escaping as volatiles, and that this was so for apples producing volatiles at relatively fast as well as at slow rates.

Since the practical application of the present research would be in the reduction of apple scald, and in the possible extension of the storage life of fruits, the first step was to undertake a survey of the concentrations of volatiles occurring in gas-stores containing apples. By making certain reasonable assumptions, the data enabled estimates to be made of the rate of production of the volatiles, but this aspect is deferred to a later paper in this series, describing large-scale laboratory experiments.

EXPERIMENTAL.

(*a*) *Methods of estimation.* Walls' method forms an excellent starting point, with its two decided advantages over other published methods, viz. that separation into the two convenient groups, ethylene and the non-ethylenic (or "odorous") volatiles, can be accomplished and that both can be expressed in absolute terms (mg. C).

It was found, however, in the present work that in estimations of the carbon content of acids from the volatile traps there was difficulty in obtaining constant values, although the method had previously been found to give consistent results with some pure organic compounds, e.g. citric acid. It was urgent to find a reliable method, and there was no time in which to investigate the subject comprehensively. Search of the literature relating to "wet" oxidations showed that different workers used different reagents, and an oxidizing agent was made up containing those most often used. Its composition was: 1 kg. fuming sulphuric acid (20 per cent. w./w. SO_3), 500 g. ortho phosphoric acid, 100 g. chromium trioxide, and 20 g. copper sulphate. The mixture was heated to 140° C. before use and was kept in stoppered bottles, with dust covers over the stoppers. Complete oxidation of such varied organic compounds as citric acid, acetic acid, and phthallic acid resulted and consistent values were obtained from the "trap" acids.

The apparatus is all glass, with taps lubricated by phosphoric acid. It is similar to that of Raistrick and Birkinshaw but no furnace is needed. A trap of glass wool moistened with acid potassium bichromate solution is introduced between the reaction flask and the CO_2 absorbent, to remove oxides of sulphur. In estimating the carbon content of a "trap" acid from a gas-store, 20 ml. of the acid are introduced into the reaction flask and 25 ml. of the oxidizing solution are run in from a dropping funnel. The flask is heated over a microburner. Evolution of CO_2 stops after about ten minutes, after which a slow stream of CO_2-free air is drawn through the apparatus for a further twenty minutes, the gentle heating being continued for all but the last five minutes. The CO_2 is absorbed in caustic soda solution in a Dreschel bottle fitted with a sintered glass spoon, is precipitated as carbonate by barium chloride, and is titrated after the excess hydroxide has been neutralized.

Using this method of estimation, the efficiency of the trapping methods has been investigated. Amyl acetate (redistilled) was used as the "non-ethylenic" volatile. It was introduced into an airstream at a slow constant rate from a burette, by displacement with mercury from a mechanically driven hypodermic syringe. The gas stream was bubbled through two traps (Dreschel bottles) in series, each containing 40 ml. of sulphuric acid. Results are given in Table 1.

TABLE I.

Trapping of amyl acetate vapour in sulphuric acid.

Air speed L./hour.	p.p.m. "volatiles".	mg. C. added.	mg. C. found by analysis.*	
			1st trap.	2nd trap.
10·8	89	65	72	2·0
5·8	194	108	103	1·6
10·1	85	145	135	0·9
11·0	32	88	91	1·0

* These figures include errors in reading the amyl acetate burette, and errors in estimation of carbon.

In view of the speeds actually used and the concentrations encountered in practice, it is clear that 40 ml. of sulphuric acid is an efficient trap for non-ethylenic volatiles.

Ethylene was added to an air-stream from a gas burette with a fine capillary tip, being displaced by mercury driven by winding a reservoir upwards mechanically. The traps comprised 40 ml. of sulphuric acid followed by 40 ml. of activated sulphuric acid (2 per cent. w./w. silver sulphate). Results given in Table 2, indicate negligible absorption of ethylene in plain acid and complete absorption in activated acid, at air speeds and concentrations met in practice.

TABLE II.

Trapping of ethylene in " activated acid ".

Air speed L./hour.	p.p.m. ethylene.	mg. C. added.	mg. C. found by analysis.	
			Plain acid.	"Activated" acid.
6·2	105	24	2·1	24
11·0	59	24	1·8	25
2·1	634	24	2·1	23
4·8	552	34	2·2	33

There is some further evidence that the plain acid trap absorbs all the non-ethylenic volatiles. If an activated acid, through which the gas from an apple store has passed (after passing plain acid) and the carbon content of which is known, is diluted with water and oxidized by potassium bichromate, it reduces the theoretical weight of bichromate, and yields the theoretical quantity of acetic acid, satisfying the equations :

(i) $C_2 H_4 + H_2 SO_4 \longrightarrow C_2 H_5 . H . SO_4 .$

(ii) $C_2 H_5 . H . SO_4 + H_2O \longrightarrow C_2 H_5 OH + H_2 SO_4 .$

(iii) $C_2 H_5 OH + O_2 \longrightarrow CH_3 . COOH + H_2O .$

Further, equations (i) and (ii) have been verified for activated trap acids from apple stores, by dilution and distillation. The fraction distilling below 84° C. gave a derivative with 3 : 5 dinitro benzoyl chloride having a m.p. of 90·5 ° C. The mixed m.p., with a derivative prepared from ethyl alcohol, was 90° C.

(*b*) *Experimental procedure.* Each gas store used in this survey had a fine ($\frac{1}{8}$ in. bore) copper tube introduced through one of the thermometer seals and reaching to about the centre of the storage space. The tubes from the different stores were led to a bench where each was connected to a Venturi flowmeter, then to two Dreschel bottles in series, and finally to a device for controlling the rate of suction of gas from the chamber. The first Dreschel bottle contained plain sulphuric acid, and the second " activated " acid.

The withdrawal of gas from the chambers was started as soon as they had been sealed. The acids in the traps were changed, each 7 to 8 days for the first few weeks of storage, and each 10 to 14 days after the 50th to 60th day. The rate of suction was recorded daily so that a reliable estimate could be made of the volume of gas which had passed through the traps. The rate of withdrawal of the gas was about 3 to 5 L./Hr., on the average.

RESULTS.

(*a*) *Units.* Rates of production of the volatiles are not considered in the present paper and indeed the data could only be made to yield such information by making certain assumptions. Rates will be discussed in later papers in this series but nowhere is it intended to adopt the unit at present in use, i.e. " mg. CO_2 " (Kidd and West, 1938a ; Potter and Griffiths, 1946), for expressing the mass of the volatiles. It is proposed to use " mg. C " as the basic unit. Concentrations of the volatiles are expressed as " mg. C per 100 litres of gas ". In the case of ethylene this may be converted to " parts per million " by volume of ethylene (by multiplying by 9·3). By making assumptions regarding the nature of the volatiles*, based on previous work (see introduction), the factor of 3·0 has been obtained for conversion of " mg. C/100 L. gas " to " p.p.m.", for the non-ethylenic volatiles. Thus a rough comparison may be made of the concentration of ethylene with that of the non-ethylenic fractions.

(*b*) *Data obtained.* Three varieties of apple have been studied and the total number of gas stores investigated total eight, in two storage seasons. Representative graphs of the concentration of ethylene and of non-ethylenic volatiles are given in Fig. 1, where the concentrations are expressed as " mg. C/100 L. gas ", and as " p.p.m. ". All stores at Canterbury were cooled by direct expansion coils in central cooling towers. The Ditton Laboratory store was cooled by brine coils, also in a central tower.

(i) Graph A : Bramley's Seedlings ; oiled paper wrappers ; 30 ton store at Ditton Laboratory, 22 x. 45 to 5 ii. 46 ; 8 per cent. CO_2, 40° F.

Ethylene fell from 14·6 mg. C/100 L. for the first seven days, to 7·5 mg. from day 23 to 106. Non-ethylenic volatiles declined steadily from 4·7 to 1·6 mg. C/100 L. by day 35, and to 0·9 mg. by day 106.

(ii) Graph B : Bramley's Seedlings ; unwrapped, 50 ton store, Canterbury ; fruit picked 30 ix. 46, loaded into store 9. x. 46, stored until 20 i. 47 ; 7 per cent. CO_2 maintained after 13th day ; temperature at loading 53° F., falling to 40° F. by 5th day.

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* Assuming that the non-ethylenic fractions contain compounds such as ethyl alcohol, acetaldehyde, and amyl esters of formic, acetic, valeric acids, then the mean carbon content is 62 per cent., and the mean molecular weight 116. This gives 1 mg. C. equivalent to 0·3 ml. vapour, or 1 mg. C/100 L.=3 p.p.m.

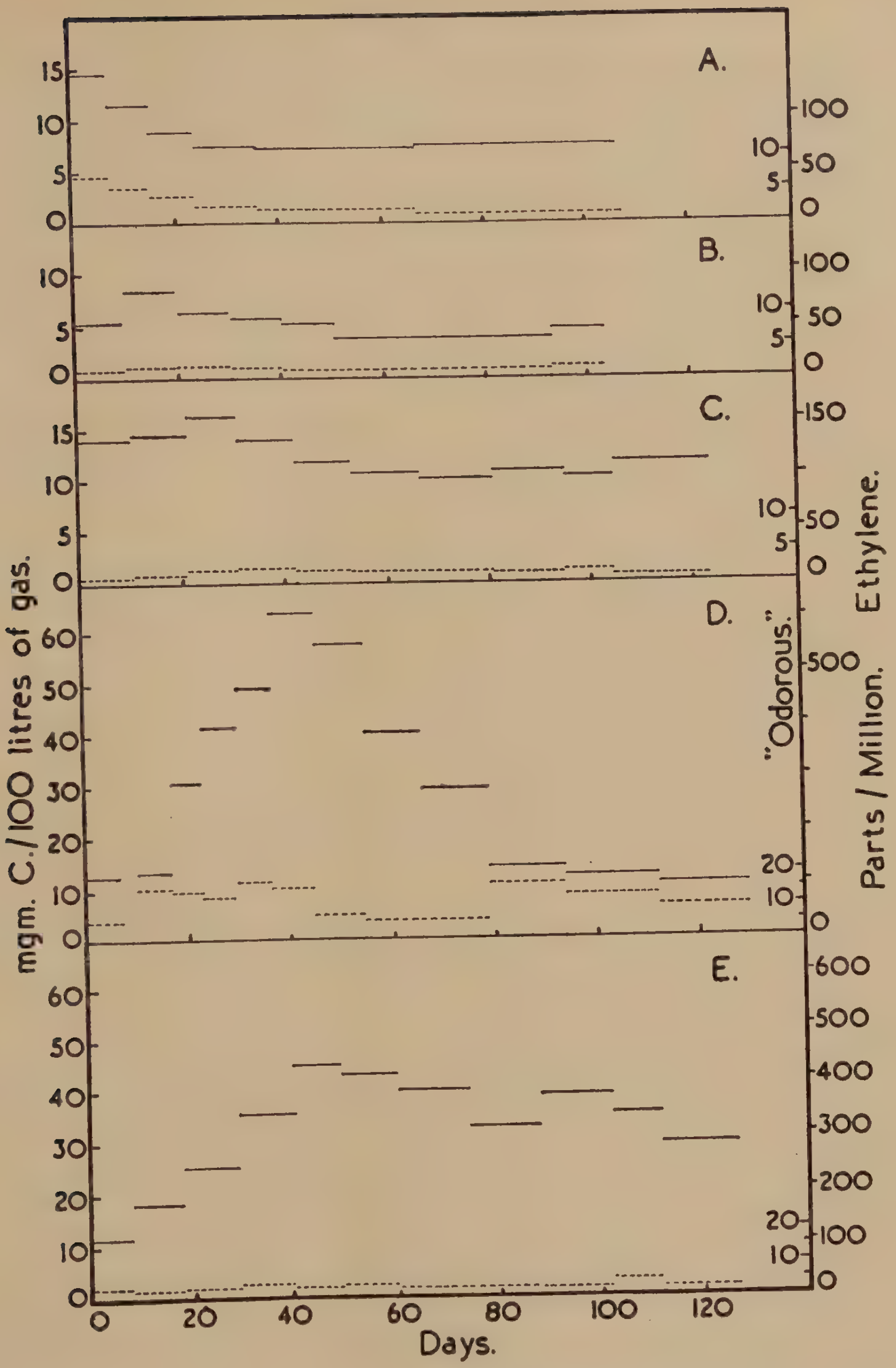

FIG. 1.

The concentration of ethylene and of non-ethylenic (" odorous ") volatiles in gas stores. (For key to graphs, see text, " Data obtained ".) Solid lines—ethylene, broken lines—non-ethylenic volatiles.

Ethylene fell from 8·7 mg. C/100 L. (the value attained when the gas concentration had risen to 7 per cent.) to 4 mg. by day 51, remaining at 4 to 5 mg. until the store was opened on day 103. Non-ethylenic volatiles remained fairly steady throughout at the low value 0·8 to 1·3 mg. C/100 L. gas.

(iii) Graph C: Lane's Prince Albert; unwrapped; 50 ton store, Canterbury; picked 24 ix. 46, loaded 7 x. 46, stored until 8 ii. 47; 7 per cent. CO_2 maintained after 10th day, temperature fell from 55° F. to 40° F. by 6th day.

The graph for concentration of ethylene shows a slight peak at 16·5 mg. C/100 L. gas on days 22 to 32, falling slowly thereafter to 10·7 mg. by day 53, and remaining at 9 to 11 mg. until day 124. The graph for the non-ethylenic volatiles tends to follow the same course, at a much lower level, the peak value being 1·4 mg. C/100 L., and the mean value about 1·0 mg.

(iv) Graph D: Cox's Orange Pippin; oiled paper wrappers; 25 ton store with "scrubber",* Canterbury; 17 ix. 45 to 25 i. 46; 5 per cent. CO_2 by 2nd day, 5 per cent. CO_2 + 5 per cent. O_2 by 7th day, ultimately falling to 5 per cent. CO_2 + 2½ per cent. O_2, loaded at 65° F., falling to 40° F. by 13th day.

Ethylene remained at 13 mg. C/100 L. until day 17, when a rapid increase in the concentration set in, the peak value of 64·2 mg. being attained from days 37 to 46. The concentration of ethylene then fell almost as rapidly as it had risen, to a value of 14·2 mg. on day 79, and to 12 to 10 mg. from days 94 to 130. Non-ethylenic volatiles showed a similar trend to begin with, having a peak value of 11·7 mg. on days 30 to 37, falling later to about 4·0 mg. The estimate for the period from day 79 to day 94 is doubtful, but the level from days 94 to 130, at 6·0 to 8·5 mg., was about twice that for the previous low period.

(v) Graph E: Cox's Orange Pippin; oiled paper wrappers; 25 ton store with scrubber, Canterbury; picked 16 ix. 46, stored 30 ix. 46 to 4 ii. 47; 5 per cent. CO_2 in 18 hours, 5 per cent. CO_2 + 2½ per cent. O_2 in 5 days, loaded at 60° F., falling to 38° F. by 11th day.

The ethylene graph shows a rapid rise to a peak at 45·1 mg. C/100 L. gas from day 39 to day 49. The fall in concentration was less rapid than in the previous year, having declined to 29·5 mg. by the 127th day. There was no evidence of a peak in the concentration of the non-ethylenic volatiles, their concentration remaining at about 2 mg. C (1·6 to 2·8)/100 L. gas for the duration of storage.

In the season 1945-46 two 50 ton stores containing unwrapped Bramley's Seedling apples in 8 per cent. CO_2 were also investigated at Canterbury. The results were similar for the two stores, with non-ethylenic volatiles practically the same as in Graph B. Ethylene gave a peak value of 14 mg. C/100 L. on days 50 to 60, thereafter declining to 9 mg. by day 110.

A 25 ton store holding Lane's Prince Albert apples, in oiled paper wrappers in 7% CO_2, at Canterbury in 1946-47 contained 10 mg. ethylene C/100 L. throughout the storage period of 107 days. Non-ethylenic volatiles had a value of 15·7 mg. during the initial cooling period, falling to 2 mg. by day 20, and running at about 3 mg. thereafter.

There exist other data, as yet unpublished, obtained by Walls in 1939-40, from gas stores at Risby, Suffolk. Walls absorbed the CO_2 in the gas drawn from the stores in soda lime, and then estimated the CO_2 produced by oxidation of the volatiles in a furnace. He has stated (1942) that soda lime absorbs part of the volatiles, and Potter and Griffiths (1946) have shown that the fraction absorbed is non-ethylenic. Because of the large amounts of CO_2 to be removed from the gas from the gas stores, Walls had to use large amounts of soda lime, and it is thus probably that the values he obtained represent ethylene concentrations.

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* The "scrubber", through which the atmosphere of the store was circulated in order to maintain a concentration of 5 per cent. of CO_2, contained a solution of sodium hydroxide in water.

His data for Bramley's Seedling, in 7 to 8 per cent. CO_2, show peak values of 25 to 30 mg. C/100 L., or about twice the highest value obtained with the Kentish apples in the present work.

In two " leaky " scrubbed gas stores containing Cox's Orange Pippin apples, with 5 per cent. CO_2 and 8 to 10 per cent. O_2, Walls found the peak value for " volatile" carbon concentration to be 30 to 36 mg./100 L. In a third store, in which the gas concentrations were 5 per cent. CO_2 + 3 per cent. O_2, the peak value was 70 to 80 mg./100 L. This value is of the same order as the peak of graph D, but the later progress of his curve of concentration of volatiles resembles the final phase of graph E.

Allington Pippin (5 per cent. CO_2 ; 4 to 8 per cent. O_2), with a peak value of 36 to 40 mg., and Laxton's Superb (8 to 9 per cent CO_2 ; 8 to 10 per cent. O_2), peak value 55 mg., appear to fall in line, as regards concentration of volatiles, with Cox's Orange Pippin apples.

It is of interest to note here that in experiments now in progress with King Edward VII apples (to be described fully in a later paper) the maximum value for ethylene carbon is of the order of 14 mg./100 L. gas.

DISCUSSION.

The first fact emerging from study of the above data is that the concentration of ethylene in a gas store containing apples always greatly exceeds the concentration of the other organic volatiles. Whereas in most of the stores the concentration of non-ethylenic volatiles was about 2 to 4 mg. C/100 L., with peak values of the order of 10 mg., ethylene carbon rarely fell below about 10 mg., rising (in the case of Cox's Orange Pippin) to about 60 mg. C/100 L.

Reference to Fig. 1 shows that if the above expressions for mass of carbon per unit volume of gas are converted to concentrations of the vapour of the volatiles, in p.p.m., the greater concentration of ethylene may be better realized. In p.p.m. the concentration of ethylene is some ten to fifty times that of the non-ethylenic fraction.

Secondly, it would appear that the dessert varieties produce more volatiles than the cooking varieties. Thus, peak values for dessert apples were : Cox's Orange Pippin, 60 mg., Laxton's Superb, 55 mg., and Allington Pippin, 35 to 40 mg. C/100 L. gas.* Corresponding values for cooking apples were : Bramley's Seedling, 14 to 30 mg., Lane's Prince Albert, 16 mg., and King Edward VII, 14 mg. C/100 L. gas.

The data suggest that, as in other respects, apples of the same variety from different districts behave differently. Thus the production of ethylene by the Kentish Bramley's gave a maximum concentration of 14 mg. C/100 L., whereas the store containing East Anglian Bramley's had twice this content of ethylene.

In one of Wall's (unpublished) experiments, he drew the gas samples from the inside of one of the apple boxes. He obtained the same values for " volatile " carbon as were given by gas samples from the general storage atmosphere. It is thus probable that, excepting possibly for a very shallow zone around each fruit, the concentration of volatiles or at any rate of the ethylene fraction of the volatiles, is uniform within the store. In the process of ventilating the gas store, volatiles will leak out as well as CO_2. The ratio of non-ethylenic volatiles to ethylene in the storage atmosphere is a measure of their relative rates of production. Thus, in gas storage at 30° to 40° F., apples produce much more ethylene than other volatiles. In the case of Cox's Orange Pippin, the production of ethylene may be as much as twenty times that of non-ethylene volatiles (in terms of carbon). In culinary varieties the ratio of ethylenic carbon : non-ethylenic carbon ranges from 4 : 1 to about 10 : 1. This ratio is very much higher than is obtained in air at higher temperatures. For example, Walls (l.c.) obtained ratios of 1·5 : 1 for King Edward VII and Laxton's Superb apples at 60° F., and Potter and Griffiths (l.c.) report a ratio of 1 : 2 for Bramley's Seedling apples in air "at laboratory temperatures". The present experiments were not designed to measure rates of production; this question of relative rates will be returned to in a later communication.

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* It may be accidental that the order of strength of the aroma of these varieties is the same as that of volatile concentration in their storage atmosphere.

If the convention of converting values expressed as "mg. C" into "p.p.m." is adopted, it will be seen that the ethylene : non-ethylenic volatiles ratio found by Potter and Griffiths becomes approximately 1·5 : 1.

The carbon lost as volatiles is only a small fraction of the total loss of carbon. In the stores containing Bramley's Seedling apples, the concentration of CO_2 was about 8 per cent. This is equivalent to 4·4 g. C/100 L. gas, or about 350 times the carbon lost as volatiles.

Kidd and West (1937) have observed that the concentrations of CO_2 encountered in gas storage suppress the climacteric rise of respiratory activity. In the light of this observation the fact that such high concentrations of ethylene are found in gas stores is all the more striking. The mean value for the culinary varieties was about 100 p.p.m., while in the store containing the Cox's Orange Pippin variety the peak value was about 600 p.p.m. These concentrations rapidly induce the climacteric in air. This point will be returned to in a later communication, describing the experiments in which the rate of production of CO_2, as well as of the volatiles, was estimated.

It is not possible at this juncture to speculate profitably on the relationship between concentration of volatiles and incidence of scald. The Bramley's Seedling and Lane's Prince Albert varieties which are susceptible to scald, produce less volatiles than the Cox's Orange Pippin, the susceptibility of which to scald is extremely slight. Nothing as yet is known either about the relative rates of production, or of the exact nature of the different fractions of the non-ethylenic volatiles; it would therefore be unjustifiable to attribute differences in susceptibility to varietal resistance factors.

The magnitude of the tasks of determining the nature of the volatiles and of their removal from the storage atmosphere, can now be assessed. The former appears to be formidable; the removal of volatiles and the effect of doing so is at present under investigation.

SUMMARY.

1. The concentration of ethylene and of non-ethylenic or "odorous" volatiles in gas stores containing different varieties of apples has been determined throughout the period of storage, and is presented graphically.

2. The concentration of ethylene exceeds, often greatly, the concentration of the odorous fraction.
3. Dessert varieties produce more ethylene than do culinary varieties.
4. The loss of carbon from the fruit, in the form of volatile organic compounds, is of the order of 0·3 per cent. of that lost as CO_2.

ACKNOWLEDGMENTS.

The author wishes to acknowledge, with thanks, the assistance given to this work by Messrs. Geo. Mount & Sons, The Nurseries, Canterbury, who have put their gas stores at our disposal, and whose foreman attended efficiently to the apparatus. Thanks are also due to Dr. L. P. Walls, formerly of the Low Temperature Research Station, Cambridge, for permission to quote from unpublished data.

The work formed part of the programme of the Food Investigation Organization of the Department of Scientific and Industrial Research.

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(*Received* 16/4/48.)

THE ORDER AND PERIOD OF BLOSSOMING IN SWEET CHERRY VARIETIES

By J. B. DUGGAN

Advisory Officer in Horticulture, National Agricultural Advisory Service, Hereford*

The order and period of blossoming of 26 varieties of sweet cherries growing in or near Sittingbourne, Kent, were recorded during the four years 1938-41. The trees concerning which the records were made each year, were growing in twelve commercial orchards and, with the exception of those in one orchard, all the trees used were mature, i.e. over 35 years old. The same trees were used each year. The orchards lay in an area 2 miles north and 2 miles south of the main road from Sittingbourne to Teynham, which are 3 miles apart, and were therefore in proximity one to another.

Although the rootstocks on which the trees were grafted are not known with certainty, it is reasonable to assume that all the rootstocks were collected as young seedling trees from local woods and would be those now known as the grey- or the brown-barked wild gaskain or mazzard. It is known that certain of the orchards recorded were planted with seedling mazzard rootstocks from the woods and that the rootstocks were subsequently grafted in the orchard with suitable scion varieties. Trees of the same variety in the same orchard were very uniform in shape, size, and in the order and period of blossoming, and although it is known that rootstock has an effect upon the flowering time of apple varieties worked onto different apple rootstocks, no such effect on the flowering time of sweet cherries is known to the writer.

Throughout the orchards the soil consisted of brick earth (very fine sandy clay loam), of a varying depth, overlying Thanet Beds material which in some cases showed mottling indicating slight impedance to the drainage of soil water. Despite the variation of the depth of the brick earth deposit, no differences in time or period of blossoming were observed which could be attributed to the soil. Differences associated with altitude were noticed ; the orchards situated on higher ground were later in flowering than on low lying ground.

The variety Adam's Crown was included for one year as it was the earliest variety to bloom in the district but it is no longer planted and will soon be lost to cultivation as the only trees existing are now very old. Hertfordshire Black was said by some fruit growers to be a suitable pollinator for Early Rivers and so was included. The varieties Cooper's Black and Mercer's Black are local varieties, but the former is likely to be planted more extensively as it is a late black cherry with good commercial qualities.

It soon became evident that the names given by the fruit growers to certain varieties of sweet cherries in their orchards were not always correct. The nomenclature of sweet cherries is still very confused and many locally raised and locally named varieties exist. Certain cherry varieties are notoriously difficult to distinguish when in fruit and characteristics such as tree shape, the size, shape and colour of leaves, period of blossoming, time of appearance of leaves in relation to flowers, have all to be used to distinguish some varieties. Coupled with this is the use of a different name for the same variety in different parts of the country. For example the Waterloo of Kent is known in Buckinghamshire as Black Eagle and the Ohio Beauty of Kent is aptly named Ironsides in Herefordshire and August Heart in parts of Buckinghamshire. Any of the varieties recorded which were thought to be incorrectly named were compared with the standard variety collections at East Malling Research Station and the John Innes Horticultural Institution and, with the exception of Hertfordshire Black and Black Cluster, as far as is known to the writer all the varieties on which records were made are correctly named. It is thought that Hertfordshire Black may well be known in other districts as Caroon. So far 3 distinct varieties are found under this name (1). Similarly there are more than one kind of Black Cluster. It would be wise, therefore, in the present account, to distinguish these two varieties as follows: HERTFORDSHIRE BLACK = CAROON (early flowering type) and BLACK CLUSTER = BLACK CLUSTER (early flowering type).

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* The work described in this paper was carried out when the author was an Assistant Adviser in Commercial Fruit Growing (Kent Education Committee).

Three different varieties of MONSTRUEUSE DE MEZEL (known more commonly as MEZEL) are listed by Crane (2). The variety recorded here as BIGARREAU DE MEZEL (1) is a white cherry and has been identified by Crane as MEZEL I.

TIME AND PERIOD OF FLOWERING

The time and period of blossoming in each of the four years is shown in Fig. 1 where the date of full flower is indicated by a cross.

In three out of the four years of recording, the dates of opening of the first flower were very close but in 1938 this date was approximately four weeks earlier. Thus a wide variation in the commencement of blossoming is shown to be possible. In 1938 the opening of the first flower was on March 16th, in 1939 on April 8th, in 1940 on April 11th, and in 1941 on April 15th.

Variation in the duration of the flowering period was very marked. It is a generally held belief that a short flowering period is followed by a heavy set of fruit. Support is given to this by the heavy crop which followed the short blossoming period of 1940 and by the small crop following the longer blossoming period of 1938. The following is the average number of days between the date of recording the first flower to open and that of petal fall for each of the four years for all varieties recorded: 1938, 22·5 days; 1939, 17·1 days; 1940, 12·1 days; 1941, 21·7 days.

The mean of the four years' records of flowering times is shown in Table I.

In the first column of the table the day numbers are based upon full bloom. Whilst these are average figures of the four years of records, the relative order of blooming of varieties is very consistent in each year. Small variations did occur, e.g. in the four years Early Rivers was in full bloom on the 8th, 5th, 4th and 7th days after the full bloom of the earliest variety recorded. The difference between 4th and 8th should be considered along with the average number of days (see above) between first flower to open and petal fall as it was noticed that in years of prolonged flowering periods there was also a prolongation of the state of coming into full bloom as well as of length of blossoming in individual varieties.

TABLE I.

Cherry varieties in order of time of coming into full bloom.

Order of full bloom in days.	Variety.
1	Adam's Crown (*1 year only*).
	Black Cluster (early flowering type).
4	Malling Black Eagle.
5	Hertfordshire Black =Caroon (early flowering type).
6	Early Rivers.
	Turkey Heart (*average of 3 years*).
7	Nutberry Black.
	Noir de Guben (*average of 3 years*).
8	Victoria Black (*average of 3 years*).
9	Governor Wood.
	Waterloo.
	Circassian.
	Bigarreau de Mezel (1) (*average of 3 years*).
10	Roundel Heart.
	Early Amber.
11	Frogmore Early.
	Bigarreau Napoleon.
12	Emperor Francis.
	Bigarreau Kentish (Bigarreau Amber).
13	Ohio Beauty.
14	Bigarreau Gaucher.
	Florence.
15	Cooper's Black.
	Noble.
19	Bradbourne Black.
20	Mercer's Black (*average of 3 years*).

SUMMARY.

1. The order and the length of blossoming of 26 varieties of sweet cherries recorded during the four seasons 1938-1941 are given.

2. The length of the blossoming period varied during the four years from twelve days to twenty-two days.

3. The order of flowering of the varieties recorded was very consistent.

ACKNOWLEDGMENT.

The recording of flowering dates of sweet cherries was undertaken at the suggestion of W. G. Kent, N.D.H., at that time Adviser in Commercial Fruit Growing, (Kent Education Committee.)

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(*Received* 1/5/48.)

NOTES ON THE VEGETATIVE PROPAGATION OF CACAO BY CUTTINGS

By D. A. RICHARDS

Formerly Horticulturist at the West African Cacao Research Institute, Tafo, Gold Coast

Many investigations at the West African Cacao Research Institute require the production of uniform clones of cacao. Although Amelonado, which is the name adopted for the most predominant variety of cacao in West Africa, appears to be genetically uniform, it has been considered advisable to use self-rooted clones for such detailed work as the definition of virus symptomatology. Also many of the Institute's disease and pest resistant selections are known to be heterozygous types of cacao, and investigations on these must of necessity be carried out on clonal material. Cacao can be rapidly propagated by budding, but this method introduces the variability associated with seedling rootstocks.

The object of this paper is to record the results obtained from the use of growth promoting substances on the root initiation and growth of cacao cuttings and to describe certain technical aspects in connection with types of cutting material.

A method of rooting cacao cuttings has been evolved at the Imperial College of Tropical Agriculture (1,4) and essentially the same technique was employed in the experiments recorded in this paper. Concrete propagating bins, filled with a coarse grade of river sand as a propagating medium and covered with glass lids shaded with palm matting overhead, were used. The material taken for cuttings was from recently hardened flush growth and was firm and woody at the base. Chupon cuttings, from terminal growths producing an erect tree, were scarce and fan cuttings, producing a low and spreading tree, usually had to be used. The lower leaves of each cutting were removed or reduced in length to avoid contact with the rooting medium and, before setting, the base of each cutting was trimmed with a sharp knife. The length of each cutting was between 8 and 12 in. and the position of the final cut did not appear to affect the rooting. The propagating bins were watered regularly three times a day to maintain the humidity and, after about one month, the cuttings were lifted and examined. After recording the number of rooted cuttings and the root length of each, they were potted into baskets and placed in a hardening bin covered with calico. In a fortnight they were moved to a shaded place to await transplanting to the field.

GROWTH PROMOTING SUBSTANCES.

During the past decade, much work has been done on the application of growth substances for the initiation of roots on cuttings, and several new methods have been evolved. The general trend has been toward the use of dry applications ; the different methods used are described in this paper. All the following investigations were made to find out the most effective growth substance and the most economic method of application. In many of the experiments it was not possible to use the same clone throughout, and the results were further complicated by seasonal differences which affected the production of roots. The data for rooting and root growth have been examined statistically and variance due to the use of different clones—usually significant—has been removed. The number of cuttings used in each treatment was 105, except where otherwise shown in the tables.

METHODS OF APPLICATION.

In each of the following methods of application, the growth substance was first dissolved in sufficient alcohol before being made up to the necessary strength with tap water or other diluent. Further information on the preparation of solutions is given by Pearse (8) and by Cooper and Stoutemyer (3).

Dilute solution dips were applied to cuttings which were first bundled together and then stood one inch deep in the solution for about 22 hours.

Concentrated solution dips were first used in 1939 by Hitchcock and Zimmerman (5), who suggested strengths from ten to one thousand times those of the dilute dips. Cooper (2) made further experiments and recommended twenty-five times the strength used for the dilute. The cuttings were dipped individually in this solution for about one second only and set at once. There was little difference between the costs of the concentrated and dilute methods.

Lanolin paste was made by dissolving 1 per cent. of the growth substance in alcohol and stirring it into the lanolin.

Talc dust was mixed with growth substances by the method recommended by Stoutemyer in 1939 (11). These instructions were carefully followed but the resulting dust was too coarse for easy application. This was overcome by rubbing the dust through a forty mesh wire sieve.

MATERIALS.

The material for cuttings was taken from selected high yielding clones of Amelonado and Trinitario types of cacao.

The growth substances used were as follows:

Hortomone A, a proprietary product prepared by Plant Protection Ltd.
β-indolyl-butyric acid.
Potassium β-indolyl-butyrate.
β-indolyl-acetic acid.
Phenyl-acetic acid.
β-phenyl-propionic acid.
A mixture of β-naphthalene-acetic acid, β-indolyl-butyric acid and phenyl-acetic acid.

Alpha-naphthalene-acetic acid and its potassium salt were not available and could not be tested.

Hortomone A. The dilute dip did not produce any striking effects and the mean change in root length was negative. The concentrated dips used were five in number, the most dilute containing 7 millilitres of the solution to 100 millilitres of tap water. This was about seventeen times the concentration of the dilute dip and appeared to give better results, but neither percentage rooted nor root length showed any significant increase over the control. The next concentration of 9 millilitres per 100 millilitres gave poor results for the percentage rooted but better for root length. Ten millilitres per 100 millilitres showed a negligible increase in the percentage rooted but gave the greatest percentage increase of root length, significant at 1 per cent., over the control in these experiments. A dip of 12 millilitres per 100 millilitres gave the highest result in percentage rooted but was inferior in root length to the previous treatment, although higher than in other treatments with this preparation. The highest concentration used gave results similar to those of the lowest concentration. It will be noticed that these results are very erratic and, although they include the highest proportionate increases both in percentage rooted and of root length observed in any of the experiments, some of the smallest are also recorded.

Table I.

Effects of Hortomone A.

Type of Application.	Concentration.	Percentage Rooted.		Root Length (inches).	
		Mean, treated.	Mean, control.	Mean, treated.	Mean, control.
Dilute Dip	0·4 ml./100 ml.	59·1	54·5	2·42	3·81
Concentrated Dip ..	7 ml./100 ml.	68·9	61·3	2·86	2·34
,, ,,	9 ml./100 ml.	67·6	64·5	4·98	3·72
,, ,,	10 ml./100 ml.	60·3	62·0	4·65*	1·70
,, ,,	12 ml./100 ml.	58·9*	30·7	3·26	2·46
,, ,,	15 ml./100 ml.	68·9	62·3	5·77	4·67

* Significant difference at 1·0 per cent.

β-indolyl-butyric acid. This had previously been used on cacao by Jordan (7) and by Cooper (2). In the present experiments, it gave good results, which were more consistent than were those of Hortomone A. The dilute solution dip was made up to the same concentration as that recommended by Jordan (7) (0·03 gm. per litre). This gave an appreciable increase in root length but not in the number rooted. Three strengths of the concentrated dip were tested. The first (0·06 gm. per 100 ml.), which was about twenty times as strong as the dilute dip, seemed to be too weak. The second concentrated dip (0·1 gm. per 100 ml.) gave not only a significant increase in percentage rooted but also an increase in root length similar to that with the dilute. The third (0·2 gm. per 100 ml.) gave good results both in percentage rooted and in root length, but only the latter effect was significant. The lanolin, with one per cent. of the growth substance in it, gave poor results. Talc dust, with the same concentration, gave excellent results in spite of the dust used being rather too coarse. In later experiments, when the talc dust was finely ground, it was found that this concentration was too high. Both the percentage rooted and root length showed significant increases.

Table II.

Effects of Indolyl-butyric acid.

Type of Application.	Concentration.	Percentage Rooted.		Root Length (inches).	
		Mean, treated.	Mean, control.	Mean, treated.	Mean, control.
Dilute Dip	0·003 gm./100 ml.	78·1	80·0	4·34	2·55
Concentrated Dip ..	0·06 gm./100 ml.	73·5	68·4	3·32	3·13
,, ,,	0·1 gm./100 ml.†	71·1*	50·3	4·24	3·54
,, ,, ...	0·2 gm./100 ml.	73·5	61·3	7·48*	3·14
Lanolin	1%	86·1	77·4	7·40	6·15
Talc dust..	1%	97·8*	71·7	14·00*	6·81

* Significant difference at 1·0 per cent.
† 210 cuttings treated.

Potassium β-indolyl-butyrate. This was used extensively by Hitchcock and Zimmerman (5, 6, 13), and has been considered by several investigators to be more satisfactory than indolyl-butyric acid, an opinion which seems to be fully borne out by the results obtained in the present experiments. The treatments were six in number, including three methods of application. The dilute dip (0·03 gm./1) gave excellent results. There was a good increase in root length and the number rooted showed an increase over the control but only the former was statistically significant. The roots were very evenly distributed around the base of the cuttings. The concentrated dips were four in number and the same range of concentrations used as those of the acid. The first (0·06 gm./100 ml.) gave good results but only the effect on percentage rooted was significant. The second concentration (0·09 gm./100 ml.) gave a good increase in percentage rooted but only a small increase in root length. The third (0·12 gm./100 ml.) showed a slight increase in percentage rooted but a considerable and highly significant increase in root length. The fourth and highest concentration (0·2 gm./100 ml.) gave very good results which appeared to be comparable to those of the same concentration of indolyl-butyric acid. Both percentage rooted and root length were significant. In the last treatment this growth substance was mixed into talc dust at a concentration of 1 per cent. At this concentration the base of the cutting was killed, but the influence of the growth substance was so great that a host of roots were produced above the dead portion of the stem. It was noticed that the lenticels burst and the roots had swollen tips as in most other cases of too high a concentration. It is suggested that 0·5 per cent. is nearer the optimum concentration for Potassium indolyl-butyrate in talc dust.

TABLE III.

Effect of Potassium β-indolyl-butyrate.

Type of Application.	Concentration.	Percentage Rooted.		Root Length (inches).	
		Mean, treated.	Mean, control.	Mean, treated.	Mean, control.
Dilute Dip	0·003 gm./100 ml.	91·1	79·9	7·60*	3·90
Concentrated Dip ..	0·06 gm./100 ml.†	78·3*	62·9	6·07	4·67
,, ,,	0·09 gm./100 ml.	70·9*	52·0	5·97	5·46
,, ,,	0·12 gm./100 ml.	90·1	89·1	11·70*	6·07
,, ,,	0·2 gm./100 ml.	99·2*	76·0	8·71*	4·58
Talc dust	1%	81·1	73·7	5·13*	2·08

* Significant difference at 1·0 per cent.
† 210 cuttings treated.

A mixture. This was made up of equal parts of β-indolyl-butyric acid, phenyl-acetic acid and β-naphthalene-acetic acid. It was desired to use the alpha form of the third chemical but this was unobtainable in time for the experiments. The mixture was used in four treatments. The dilute dip (0·04 gm./litre) gave poor results. The first of the concentrated dips (0·05 gm./100 ml.) showed an insignificant increase over the controls. The second (0·07 gm./100 ml.) gave very promising results both in percentage rooted and in root length, the latter being highly significant. The final and highest concentration of the mixture appeared to be too strong as it gave much smaller increases than either of the other two concentrated dips.

TABLE IV.

Effects of a mixture of β-indolyl-butyric acid, phenyl-acetic acid and β-naphthalene-acetic acid.

Type of Application.	Concentration.	Percentage Rooted.		Root Length (inches).	
		Mean, treated.	Mean, control.	Mean, treated.	Mean, control.
Dilute Dip	·004 gm./100 ml.	50·0	55·9	3·74	5·50
Concentrated Dip ..	·05 gm./100 ml.	61·4	53·5	4·33	4·06
,, ,,	·07 gm./100 ml.	90·6	80·3	3·75*	1·99
,, ,,	·1 gm./100 ml.	80·0	79·9	3·46	2·84

* Significant difference at 1·0 per cent.

β-indolyl-acetic acid. A concentrated dip at 0·1 gm. per 100 ml. was tested and gave poor results. This may have been due to too low a concentration but it was not tried further owing to the satisfactory results of the butyric compounds.

Phenyl-acetic acid. This gave insignificant results at 0·3 gm./100 ml. It was of interest on grounds of cost as it is one of the cheapest chemical growth substances.

β-phenyl-propionic acid. At 0·2 gm./100 ml. this was the final growth substance to be tried. It gave an increase in number rooted, as with the two previous chemicals, but failed to give an increase in root length.

Only one concentrated dip of each of these three substances was tried and the results are grouped in Table V.

TABLE V.

Effects with three acids.

Acid used (concentrated dip).	Concentration.	Percentage Rooted.		Root Length (inches).	
		Mean, treated.	Mean, control.	Mean, treated.	Mean, control.
Indolyl-acetic acid ..	0·1 gm./100 ml.	84·6	74·4	3·02	3·24
Phenyl-acetic acid ..	0·3 gm./100 ml.	78·6	66·1	1·92	1·99
Phenyl-propionic acid† ..	0·2 gm./100 ml.	67·1	53·5	3·56	3·78

† Only 60 cuttings treated.

DISCUSSION.

The dilute solution dips were found to be slow and difficult to manage where large numbers of cuttings had to be handled. It was usually necessary to bundle cuttings before standing them in the solution for 22 hours. The use of the concentrated dips was found to be simpler as the cuttings were dipped for one second only and then set in the sand rooting medium. Both the solution methods present the difficulty that the solutions must be made up afresh from a stock bottle each time they are required. The use of talc dust as a carrier obviates this difficulty and a stock mixture can be stored and used over a period of several months if kept in a dark place.

It was found essential to use a dibbler for setting the cuttings otherwise the dust on the base of the cuttings was rubbed off by the sand. The dust, which must be very fine for satisfactory results, can be made to adhere to the base of the cutting if the latter is first moistened. Growth substances mixed with lanolin were not easy to apply and did not give good results. The most effective methods for the application of growth substances appeared to be the concentrated one-second-dip solution or in talc dust. Of the substances tested, β-indolyl-butyric acid and potassium β-indolyl-butyrate were the most consistently effective.

TYPES OF CUTTING MATERIAL

When material for propagating is scarce, as is often the case with a selected clone, and large numbers of plants are required quickly, the supply can be increased by budding onto seedling stocks. The budlings take several years to produce cutting material in any quantity and other methods were investigated to find a more economical way of propagating than the large-cutting technique already described. There was no experimental layout and few numerical data are available. The methods of propagating plants on their own roots other than by cuttings were found to be both slow and difficult besides being uneconomical of material.

Semi-hardwood cuttings. These are employed in the usual method of propagating cacao by cuttings and the technique has already been described. The method was evolved by Pyke in 1932 (10) and developed in 1935 by Cheesman and Spencer (1).

Dwarf cuttings. This name was adopted for small cuttings between two and four inches in length. Cuttings of this size were first tried using green material which was just hardening but more cuttings wilted and rotted than if made of harder wood. The rooting variation between the two main types of cacao, Trinitario and Amelonado, seemed to be more pronounced in these small cuttings than in the larger. Dwarf cuttings were, however, the most economical as regards material and usually rooted as well as the normal cuttings. They took much longer than semi-hardwood cuttings to reach a size suitable for transplanting.

Leaf-bud cuttings. These had not previously been tried with cacao, and most of the information available seemed to be from the propagation of various species of Rubus. Watkins and Blackmon (12) did some work on the vegetative propagation of various tropical plants by means of leaf-bud cuttings. In the investigations at Tafo, the cuttings used consisted of a half-inch section of stem with a leaf attached ; part of the leaf was removed to reduce the risk of disturbance when watering. It was preferable to have a prominent axillary bud on each cutting or the bud might fail to develop after rooting. Only 20 per cent. of these cuttings rooted and both the hardening-off and later growth were difficult and slow. Another method tried was to take leaf-bud slips in the same way as used in budding but with a leaf attached. They were set in fine sand under a bell-jar. Some of these rooted after six or seven weeks but the procedure was even more difficult than with the previous method.

Root cuttings. This type was used by Pyke in 1931 (9) in his preliminary investigations in rooting cacao, but he reported that all died within a few months without any sign of either roots or shoots. The cuttings tried at Tafo were about 4 in. in length by $\frac{3}{8}$ of an inch in diameter and they were made from young wood. Those treated with 1 per cent. indolyl-butyric acid in lanolin, rooted well in about three weeks but, as before, the untreated ones all failed to root. About 70 per cent. of those treated rooted and later produced good root-systems but they failed to produce any sign of a bud and died after a few months.

Hardwood cuttings. Pyke (9) also tried hardwood cuttings from 5 to 12 in. long, of wood from vigorous fan material less than two years old and from $\frac{1}{4}$ to $\frac{3}{8}$ of an inch in diameter. Although buds grew well, all failed to root. Experiments were repeated at Tafo on similar material but about 4 in. in length and up to $\frac{1}{2}$ in. in diameter. Those treated with 1 per cent. indolyl-butyric acid in talc dust, rooted well in a month but again the untreated cuttings failed to root. Buds developed on the apex of the cutting whether it had rooted or not. Trouble arose when attempts were made to harden off these cuttings. The very young leaves failed to mature and were shed although several hardening techniques were tried. If these cuttings could be hardened off successfully, it would be most useful, as sections of chupon stem could be taken, thus producing many more erect trees.

CUTTINGS OF TWO TYPES OF CACAO.

During the first few months of the propagation of cacao by cuttings in West Africa, a great variation between the rooting of one selection and that of another was noticed. This was thought to be either an irregularity in the technique of making cuttings or else a characteristic feature of the individual selection. The standard technique was used. Later, however, it was noticed that this irregularity in rooting was correlated with the type of cacao. The two types being propagated were Trinitario and West African Amelonado. The hybrid Trinitario selections were usually more vigorous and they gave a much higher rooting percentage. This vigorous rooting did not appear to be altogether related to the visible vigour of the tree but rather to independently inherited characteristics. Thus a vigorous and healthy-looking Amelonado tree would give cuttings that would invariably root less well than cuttings from poor-growing Trinitario. Records were taken of the rooting of eight typical selections of each type. The Amelonado selections rooted from 38 per cent. to 57 per cent. with an average of 48 per cent.; the Trinitario selections from 62 per cent. to 88 per cent. with an average of 73 per cent. This difference of 25 per cent. is significant at 0·10 per cent.

SUMMARY.

Investigations were carried out on the improvement of methods of vegetative propagation of cacao clones for experimental purposes.

1. Semi-hardwood cuttings were treated with Hortomone A, indolyl-butyric acid, potassium indolyl-butyrate, indolyl-acetic acid, phenyl-acetic acid, phenyl-propionic acid, and a mixture of growth substances, in dilute and concentrated solutions and with lanolin and in talc dust.
    - (1) Potassium indolyl-butyrate in a dilute solution gave significant increases in number rooted and in root length.
    - (2) Concentrated dips for one second using indolyl-butyric acid, potassium indolyl-butyrate, and a mixture of indolyl-butyric acid, phenyl-acetic acid and β-naphthalene-acetic acid, gave significant increases in percentage of cuttings rooted and in mean root length.

(3) Talc dust mixed with indolyl-butyric acid gave significant increases and this method was the most simple to apply.

2. Types of cutting material, including dwarf, leaf-bud, root, and hardwood cuttings were all successfully rooted but were more difficult to establish than the usual semi-hardwood type of cutting

3. Trinitario type cacao rooted significantly more easily than West African Amelonado.

ACKNOWLEDGMENTS.

I have to thank Dr. D. W. Goodall for the statistical analysis of the data and for his very helpful advice in the preparation of this paper. I am also indebted to the African Staff engaged on propagation and to the Director of Cacao Research, Mr. O. J. Voelcker, for permission to publish this paper.

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(*Received* 10/5/48.)

EXPERIMENTS ON THE LAYERING OF HOPS

I. THE ESTABLISHMENT OF A PERMANENT LAYER BED*

By W. G. KEYWORTH, MARGARET M. HITCHCOCK and DOROTHY J. WILSON
East Malling Research Station, Kent

THE growing importance of certain hop diseases, notably Verticillium Wilt, Nettlehead and Mosaic, has emphasized the need for the rapid production of special disease-free or disease-resistant stock. The normal commercial method of hop propagation involves the planting of basal cuttings either direct into the new field or into a nursery bed to produce one-year-old plants, the " rooted " or " bedded " sets of commerce. These cuttings are produced by earthing up the base of the fruiting bines (stems) on the plant, the earthed portions remaining alive after the aerial part of the bine has died back at the end of the season. When allowance has been made for some subsequent loss, it may be estimated that each mature plant thus normally produces about five new plants but there is some variation according to the number of bines trained up in the different systems of stringing. The rate of multiplication of the stock is therefore slow and, because of the large number of parent plants involved, adequate roguing for disease is often impracticable.

Studies have been made at East Malling of methods of increasing the rate of multiplication of stock. Beard (1944) described a method of layering applicable to some commercial plantations but emphasized the need for special nurseries for the purpose. Beard and Wilson (1946, 1947) investigated the use of softwood cuttings by rooting them in frames, but concluded that, although showing promise with certain varieties, the methods were unlikely to be of wide application because of the equipment and attention required. The writers have therefore studied means to improve the layering technique and a method suitable for a permanent nursery is described below. Another communication (Keyworth and Wilson, 1948) describes a modification of this method using young plants.

THE LAYERING PROCESS.

As already stated, hop bines die back to soil level at the end of each season, but those portions of the bine which are below the soil remain alive. Layering is an extension of the commercial method of earthing the base of the bine and enables a greater length of bine to be covered with soil and thus provide a number of cuttings. The portion beneath the soil thickens, probably owing to the accumulation of food reserves and produces adventitious roots and also buds at each node. At the end of the season the length of layer is cut into portions some four inches long, each containing at least one node. These cuttings are then planted in a nursery bed for one year to produce bedded sets.

Method No. 1.

This method, described by Beard (1944), has been in use for many years at the Research Station. It consists in laying a maximum of six bines per plant (surplus to

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* Part of this work was carried out with the aid of a grant from the Agricultural Research Council.

those required to produce the crop) on the ground along the row when they are some two feet long. They are then covered with soil except at the tips, which are free to grow. This covering process is repeated as the bines lengthen until the tips can climb the strings of the next plant. Although relatively simple, the method has been found in practice to have two main disadvantages, viz. the growing layer bines are at ground level and have no supporting strings, thus being easily damaged by cultivators, etc., and when they reach the next plant they have to compete with its already advanced bines.

Method No. 2.

To overcome the disadvantages of method No. 1, the following technique was devised, particularly for use in special hop nurseries.

The bines to be layered are grown up strings until they have reached a height somewhat in excess of the total layer length (i.e. the distance between adjacent plants in the row). They are then taken down, complete with their strings, and laid in a shallow trench along the row, their ends overlapping the adjacent plant. These ends of the bines are then trained up new strings put in the positions vacated by the layer bines of the adjacent plant and are thus able to grow vigorously. The layered portions can then easily be covered with a ridge of soil some nine inches high and eighteen inches wide by ploughing down each side of the row and filling in the centre of the ridge.

Special wirework not less than eleven feet high is erected and a convenient spacing for the plants is 7ft. 6 in. apart and 7-8 ft. between the rows. Up to 8 bines are selected on each mature plant, two being " feeding " bines which are unlayered and serve to nourish the rootstock, the remainder being layered. The feeding and layer bines are trained up separate pairs of strings attached at ground level to a " screw peg " and passing over hooks on the top wire. As the hop plant takes about three years to mature, fewer bines are available for layering in the first two years, but the method is still applicable. The process is illustrated in Fig. 1.

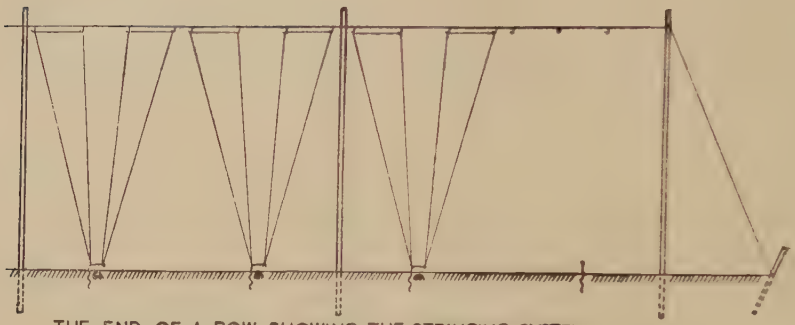
At the end of the season, the layers are lifted and the cuttings from them planted in a cutting nursery as already described by Beard (1944).

FIELD EXPERIENCE WITH METHOD NO. 2.

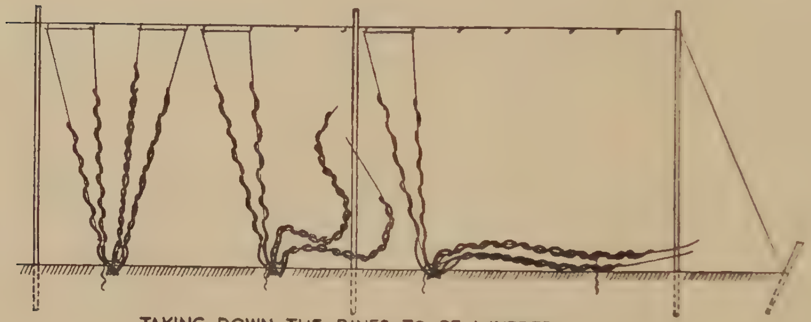
The technique described has been employed in establishing layer beds of varieties resistant to Verticillium Wilt and also clonal selections of the Fuggle variety and certain varieties of Goldings.

It was observed on these layer beds that the layered bines grew readily up their new strings and appeared invigorated by the layering process. Their leaves were generally of a darker colour than those on the unlayered bines and they bore a good crop of hops. The process of layering, involving only one operation, was also simplified and there was little danger of damage to the tips.

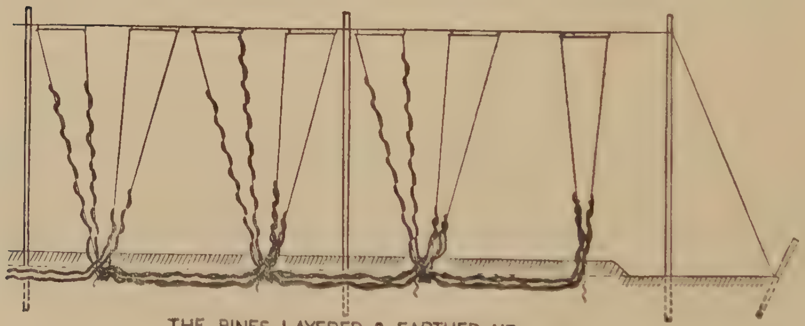
Some trials were made to discover the effect of layering the bines later for a further 7 ft. 6 in. and it was found that the ends of the bines would still grow to the tops of their new strings. This meant that the layered bines were some 27 ft. long, 15 ft. of this length being layered. It became apparent, however, that this second layering had to be done as soon as the bines were of an adequate height. If it was delayed, the bines apparently became too mature to form good layers and for this



THE END OF A ROW SHOWING THE STRINGING SYSTEM



TAKING DOWN THE BINES TO BE LAYERED



THE BINES LAYERED & EARTHED UP

FIG. 1.

Diagram of layering method No. 2.

reason, in the few trials which were made, an excessively high proportion of " blind " cuttings was obtained on the second length. This resulted in an average increase of only some three or four good cuttings per bine and the practice was discontinued until further information could be obtained on the optimum time for the second layering.

A number of practical difficulties were encountered. In the early trials the leaves were stripped from the bottom four feet of the layer bines to facilitate spraying, but this was found to damage the bud initials at the nodes and had to be discontinued. There was a slight tendency for some of the bines to break away from the rootstock when layered unless care was taken not to bend them too sharply. Uneven growth of the bines caused the tips of some of the shorter ones to be buried by the layering process with consequent death of the bine.

The layers when dug up at the end of the season were, of course, twisted round each other and the string, and care was necessary in untwisting them.

Incomplete appreciation of some of these difficulties resulted in a yield from the layer beds which was smaller than it might have been. Summaries of some of the results on two sites are given in Table I. Site 1 was on a heavy clay loam and Site 2 on a medium sandy loam.

TABLE I.

Layer bed. Site 1. (Heavy clay loam.)

Variety.	Year of layering.	Age of plants (years).	No. layered.	No. layer cuttings/plant.	No. layer cuttings/bine.
OJ47 ..	1945	1	32	33	14·8
	1946	2	35	67	16·4(1)
	1947	3	31	41	8·6
AEE55 ..	1945	2	16	67	14·2 (2)
OR55 ..	1946	1	46	32	15·4
	1947	2	94	39	10·2

Layer bed. Site 2. (Medium Sandy loam.)

Variety.	Year of layering.	Age of plants (years).	No. layered.	No. layer cuttings/plant.	No. layer cuttings/bine.
OJ47 ..	1945	1	18	18	16·2
	1946	2	33	28	9·8
AEE55 ..	1945	2	16	71	11·8
OR55 ..	1945	1	29	26	13·6 (3)
	1945	2	24	57	12·2 (3)
	1946	3	24	33	10·2

Notes.—All the plants on both sites produced an average of two basal cuttings per plant in addition to the layer cuttings.

(1) 40 out of 142 bines layered for 15 feet.
(2) 17 out of 66 bines layered for 15 feet.
(3) All bines layered for 15 feet.

The results from these commercial trials show considerable variations between sites, varieties and years of layering. Many of the variations were undoubtedly due to differences of management on the two sites and in different seasons, but a strong impression was gained that the number of cuttings per bine tended to decrease as the plants became older. This is supported to some extent by the figures given. It seems likely that bines from old plants generally have longer internodes than have those from young plants and this factor may have to be considered in assessing possible yields over a period of years.

COMPARISON OF METHODS 1 AND 2.

The two methods of layering differ fundamentally in two respects, viz., (*a*) In method No. 1 the bines are exposed for relatively short periods only (i.e. until the portion to be buried has grown some two feet), whereas in method No. 2 the bines are exposed for their whole period of growth until layering (some 9-10 weeks). (*b*) In method No. 1 the portions of the bines to be buried are young and may be expected for this reason to have shorter internodes than the bines layered by method No. 2.

A simple trial to compare the number and size of cuttings produced by each method was therefore made using duplicated plots on which each method was tested on three-year-old plants of varieties OR55 and OJ47.

The layers were lifted, counted and weighed and then divided into cuttings of standard length (about 4 in.) which were also weighed.

A summary of the results is given in Table II.

TABLE II.

Summary of records from trial to compare Methods 1 and 2.

Variety.	Layering method.	No. of plants.	No. of layers per plant.	No. of cuttings per layer.	Average weight of cutting (gm.)
OR55 ..	1	14	4·8	9·5	7·1
	2	14	5·5	9·5	9·5
OJ47 ..	1	14	5·0	8·2	6·6
	2	13	3·5	8·3	7·8

It is apparent that, under the conditions of the experiment, the two methods gave very similar average numbers of cuttings per layer. Method No. 2, however, gave significantly heavier cuttings than Method No. 1, an effect which was greater in the variety OR55 than in OJ47. It was noted that the ends of the bines layered by Method No. 2 grew much more vigorously than those layered by Method No. 1, a fact which possibly accounts for the greater weight of the cuttings.

No trials have yet been conducted on the percentage " take " of cuttings raised by each method, but field experience suggests that there is little difference. Judged solely on the basis of the number of cuttings produced, therefore, the two methods would appear to give comparable results. In the trial, however, care was taken to minimize damage to the bines layered by Method No. 1, a precaution which is difficult to observe in commercial practice.

DISCUSSION.

It is likely that each of the layering methods described will prove useful under appropriate conditions, but Method No. 2 seems to possess practical advantages when used in a special layer bed. The possibilities of damage to the bines are reduced and the layering is carried out in one operation with consequent ease of earthing the layers.

The numbers of feeding and layer bines (viz. two and up to six respectively) were decided on an empirical basis, but have proved reasonably satisfactory in practice. The retention of a certain number of unlayered feeding bines was based on the supposition that the rootstock might otherwise be starved of translocated food material. It was believed that in any bine which was layered the major portion of such food material would be stored in the layer and thus never reach the rootstock. The enhanced vigour of the layered bines, however, combined with the production of roots on the layers, suggests that the layering process results in the production of a substantial amount of extra food material. It is thus conceivable that sufficient material may be produced by layered bines both to form the layers and to enable the rootstock to grow. This supposition is supported by evidence from experiments with young plants described elsewhere (Keyworth and Wilson, 1948). Plants on which all the bines were layered, however, might form an abnormally shaped rootstock resulting eventually in a dearth of strong bine. Experiments of sufficient duration to test the growth of layered plants over a number of years or to determine whether the annual harvest of layered bine will eventually weaken the rootstock have not yet been made.

ACKNOWLEDGMENTS.

The writers are indebted to Mr. F. H. Beard for help and advice and to the Statistical Section of the Research Station for analyses of the data. The assistance of the farm staff of Messrs. Whitbread & Co. is also gratefully acknowledged.

SUMMARY.

Descriptions are given of two methods of establishing a permanent hop layer bed. One which has been in use for some years possesses certain practical disadvantages, and the other is a modification designed to eliminate these. Notes are given of practical experience with this latter method and the two methods are compared from the points of view of yield of cuttings and suitability for commercial application.

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(*Received* 14/5/48.)

EXPERIMENTS ON THE LAYERING OF HOPS

II. LAYERING YOUNG PLANTS*

By W. G. KEYWORTH and DOROTHY J. WILSON
East Malling Research Station, Kent

Attention has been drawn, in a previous paper (Keyworth *et al.*, 1948), to the need for the rapid multiplication of special stocks of disease-free or disease-resistant hops and two methods have been described for establishing a permanent layer bed for this purpose. These methods depend on the fact that any portion of the bine (stem) which is covered with soil, remains alive at the end of the season when the aerial part of the bine dies. This layered portion can be divided into cuttings (each including at least one node) which are then planted for one year in a nursery to produce " bedded sets ". During the course of work on the selection of varieties of hop resistant to Verticillium Wilt (Keyworth 1947a) it became apparent that although the new propagation methods represented an advance on normal commercial practice, the rate of multiplication of special stocks was still low. The main reason for this was that the methods were intended primarily to provide a continuous supply of stock from *mature plants*, the layer bed being designed accordingly. No plants younger than one-year-old bedded sets were used and, although some layers were produced in the first year of growth, the layer bed was not at maximum productivity until the second or third year. It was already clear that bedded sets could be layered, and probable that by concentration on this process without attempting to make a permanent bed, a saving in land and equipment could be made. Furthermore, and even more important, it seemed probable that, if the bines from *cuttings* could be layered in their first year of growth, the time lag involved in producing sets and mature plants could be eliminated with a consequent rapidly increasing annual rate of multiplication.

Means of propagating from bedded sets and cuttings were therefore studied. Hop cuttings are normally planted in a nursery bed to grow for one year to produce the one-year-old plants known as bedded sets. It is usual to allow the bines from the cuttings to grow up temporary supports, such as strings laced between wires about 4 ft. apart and supported on short poles. Preliminary tests made in 1946 showed that if these bines were laid on the ground for a portion of their length and covered with soil, those parts beneath the soil thickened and formed layers in a manner analogous to that described elsewhere for mature plants (Keyworth *et al* 1948). In addition, each surviving cutting also produced a bedded set.

In order to obtain more information on the layering of the bines both from bedded sets and from cuttings, particularly in respect of the optimum planting and layering distances, further trials were made in 1947 as described below.

Layering Method.

Two promising wilt-resistant varieties OR55 and OJ47 (Keyworth 1947b) were used in the trial, with the object of producing urgently needed stock.

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* This investigation was made with the aid of a grant from the Agricultural Research Council.

Poles 7½ ft. high in the case of the sets and 5½ ft. high for the cuttings, were spaced at intervals of 14 ft. along the rows and connected by a three-strand galvanized straining wire running the whole length of the row near the tops of the poles and firmly guyed at each end. The rows were five feet apart to allow cultivation by a small tractor, and the sets or cuttings were spaced at the requisite experimental distances along them.

To enable the bines from a large number of plants to be layered simultaneously the following method was used:

A flexible layering wire (e.g. telephone cable) was fastened to a nail in the first pole at a height of one foot from the ground and stretched over another nail in that pole just below the straining wire and six feet* above the lower nail; thence over a top nail in each pole to the end of the row where it was fastened to the top nail in the end pole. Each plant was provided with a vertical string attached to this layering wire and fastened at ground level to a " screw peg " or bottom wire. Pairs of nails ¼ in. apart were put in each pole 1 ft. from the ground with a single nail at this height in the last pole. To layer the bines the wire was unhooked from the top nails and stretched tight along the row at the height of the lower nails, being slipped between each pair of nails on each pole. The slack at the far end of the row was taken up by putting the wire under the lower nail on the last pole whence it stretched up to the top nail. The effect of this was to lay the strings and bines along the ground for six feet, the remaining foot of string and bine rising vertically to the layer wire.

This technique was adapted for any length of layering by an adjustment of the positions of the lower nails, the layer length being equal to the distance between them and the top nails.

The layered bines were then covered by ridges about 9 in. high made along the row by ploughing down each side, the centres of these ridges being filled with soil to enclose the bines completely. New strings were fastened from the lowered wire to the top wire and the bines allowed to grow freely up them.

EXPERIMENTAL DESIGN.

1. *Bedded sets.*

The effects of three planting distances (2 ft., 3 ft., and 6 ft.) and two layering distances (3 ft., and 6 ft.) were studied in randomized blocks, the six possible combinations of spacing and layer lengths being replicated four times. In order to keep the two varieties separate they were planted in complete rows running throughout the trial. Each plot was four rows wide, having one experimental row of OR55 and one of OJ47, flanked by similarly treated guard rows of OR55. There were six experimental plants of each variety in each plot and the plots thus varied in size according to the spacing of the plants. Sufficient guard plants were put at the ends of each plot to ensure that all the experimental plants in the plot were alike in respect of overlap of layers.

2. *Cuttings.*

The spacing distances for cuttings were 1 ft. and 2 ft. and the layer lengths were 2 ft. and 4 ft. Control plots of unlayered plants were also included in order to compare the sets produced with those from the layered plants. There were thus six treatments in all which were replicated six times. One block of treatments was planted with " strap cuttings " (i.e. cuttings obtained from bine bases in the normal commercial way), the others being planted with layer cuttings. End guard plants and guard rows were used in the same fashion as in the trial of bedded sets but this trial differed in having plots of equal length which thus contained twice as many cuttings at 1 ft. spacing as at 2 ft. spacing.

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* A layer length of six feet is given as an example of the general procedure (see also Figs. 1 and 2.)

EXPERIMENTAL PROCEDURE.

Most of the sets were making good growth by the end of May, only ten in the whole experiment having failed. Although bine growth was variable, by mid-June some of the bines had reached the top wire and it became necessary to select the optimum time for layering to ensure that most of the bine was as near the tops of the strings as possible without being unduly tangled with the fixed straining wire. Accordingly the experimental set-rows of OR55 were layered on 19th June, followed by the OJ47 on 27th June and the guard rows on 2nd, 3rd and 8th July.

By the end of May, the cuttings showed a " take " of about 75 per cent. Growth was very variable but by the time they were layered, in the second and third weeks of July, most of the backward bines had grown to the necessary height.

After layering, those bines which had their tips free from the soil grew vigorously and reached the top wire in a few weeks. In both trials they bore a crop of hops. These were picked from the variety OJ47 but as it was found that this caused some damage the OR55 hops were left unpicked. Owing to the almost complete absence of aphids in 1947, nicotine sprays were unnecessary, but the plants were sprayed with Bordeaux mixture in late July and early August as a protectant against Downy Mildew. Very little of this disease was seen on the plot at any time but there was a heavy infestation of Red Spider in July and August. The season was extremely dry and hot throughout and the soil was in consequence not so moist as could have been desired.

Lifting began in the set trial in mid-October, followed by the cutting trial which was completed by 1st November. The aerial portions of the bines were cut off about 6 in. above the ridges and removed. The layers were carefully pulled up and the sets lifted with spades, ensuring that similar amounts of root were lifted on each set.

EXPERIMENTAL RESULTS.

1. *Set trial.*

The records of greatest interest in this trial were, (1) the survival of the parent plants, (2) the numbers of bines layered and (3) the numbers of good and blind cuttings per survivor and per set planted. The mean values of these for each treatment are summarized in Table I.

The records on which these summaries are based were submitted to the analysis of variance and gave the following information (differences are significant at the 5 per cent. level and apply to both varieties unless otherwise stated).

(*a*) *Survival of parent plants.* Fewer plants survived when layered for 6 ft. than when layered for 3 ft. It was noted in the course of the experiment that the complete burial of a bine killed it and if all the bines on a plant were so buried the whole plant died. The lower survival rate at 6 ft. layering is presumably due to the

TABLE I.

Summarized records from set trial.

Variety.	Treatments.		Survivors (% of 24 planted).	Mean values per surviving plant.			Good cuts per Set planted.
	Layer length. (ft.)	Spacing. (ft.)		Bines layered.	Good cuts.	Blind cuts.	
OR55 ..	3	2	79	1·6	16·3	0·0	12·9
		3	88	1·8	18·6	0·3	16·2
		6	92	1·6	15·7	0·2	14·4
	6	2	67	1·4	17·3	1·7	11·5
		3	79	1·3	16·9	1·8	13·4
		6	71	1·1	13·7	2·4	9·7
OJ47 ..	3	2	96	1·7	18·3	1·1	17·5
		3	83	1·6	19·6	0·8	16·3
		6	100	1·6	18·3	0·6	18·3
	6	2	75	1·2	13·4	5·3	10·0
		3	58	1·1	13·4	5·5	7·8
		6	38	1·1	13·1	4·7	4·9

fact that although most of the plants had bines over 3 ft. long when layered, a number had none over 6 ft. long. These plants were thus killed by the burying of their bines in the 6 ft. layering process. Distance of spacing did not affect survival.

(*b*) *Numbers of bines layered.* The 6 ft. layer plots contained fewer bines per plant than the 3 ft. plots, an effect also due presumably to the death of the shorter bines on some of the plants when buried by the 6 ft. treatment.

(*c*) *Numbers of cuttings produced.* There was a slight but non-significant increase in the number of cuttings produced on the 6 ft. as compared with the 3 ft. layers. The lower survival of bines at 6 ft. layering, however, resulted in a similar number of cuttings being produced by each treatment. Furthermore, there were more blind cuttings on the longer layers (particularly in the variety OJ47) resulting in these giving actually a smaller number of plantable cuttings than the short layers. If, in addition, the survival of the parent plants is taken into account, the 3 ft. layer treatment was much superior in the production of good cuttings per set planted. A direct measure of the weights of the cuttings was not obtainable as the layers were not cut up. However, an estimate based on a standard length of cutting, showed no differences in cutting weight between treatments. All the "good" cuttings from each treatment were of plantable size.

2. *Cutting trial.*

In addition to the survival of the parent plants, and the numbers of layers and cuttings obtained, a record of interest in this trial was the effect of the treatments on the weight of set produced from the parent cuttings. The records are summarized in Table II.

TABLE II.

Summarized records from cutting trial.

Variety.	Layer length (ft.)	Spacing (ft.)	% Survivors.	Mean values per surviving plant.					Good cuts per cutting planted.
				Bines layered.	Good cuts.	Blind cuts.	Weight of layers. (gm.)	Weight of set (gm.)	
OR55	0	1	93	—	—	—	—	92·1	—
		2	83	—	—	—	—	114·0	—
	2	1	84	1·4	10·7	0·1	82·3	77·7	9·0
		2	87	1·7	12·7	0·2	108·3	96·2	11·0
	4	1	68	1·2	12·8	1·1	134·7	78·2	8·7
		2	53	1·3	14·5	0·6	182·8	97·2	7·7
OJ47	0	1	81	—	—	—	—	58·7	—
		2	82	—	—	—	—	75·3	—
	2	1	66	1·4	9·9	0·3	65·6	62·6	6·5
		2	70	1·5	10·0	0·2	82·9	77·9	7·0
	4	1	45	1·1	9·6	2·2	100·6	68·7	4·3
		2	58	1·2	10·5	1·5	112·3	82·3	6·1

An analysis of variance of the records for this trial gives the following information :—

(*a*) *Survival of parent plants and numbers of bines layered.* Both the layering treatments reduced the survival of plants of the variety OJ47, the 4 ft. treatment having the greater effect. Only the 4 ft. treatment reduced the survival of OR55, however, the 2 ft. treatment having no effect in this respect. The differences between the two varieties were probably due to the poorer growth of the variety OJ47 resulting in a greater proportion of buried bines. The 4 ft. layer treatment somewhat reduced the number of layers per surviving plant in both varieties, although the differences were not significant.

(*b*) *Numbers of cuttings produced.* The OJ47 plants layered for 4 ft. gave slightly more cuttings than those layered for 2 ft. The OR55 plants showed a similar but non-significant effect. These increases, however, were more than offset by the incidence of blind cuttings and the reduced survival of the plants when layered for four feet. The total effect of these factors was that the plots layered for 2 ft. gave much the greater number of plantable cuttings.

(*c*) *Weights of sets and layers.* One of the objects of the trial was to determine the effects of layering on the size of set produced from the parent cutting. The two varieties gave different results in this respect. The sets of the variety OR55 were slightly reduced in size when the bines were layered but still remained of good plantable size. On the other hand the sets of the variety OJ47 showed a fairly definite but non-significant increase in weight with increased layer length. An addition of layer and set weights shows that the layering process resulted in all cases in a considerable increase in the total weight of propagating material from each cutting. The more widely spaced cuttings gave bigger sets in all cases, an effect due presumably to a reduction in root competition. Fig. 3 illustrates typical examples of sets and attached layers produced by the three layer treatments (0, 2 ft. and 4 ft.).

(*d*) *Comparison of layer and strap cuttings as parent plants.* As already noted, one block was planted with strap cuttings in order to give some comparison of their performance with that of the layer cuttings comprising the remainder of the trial. (The strap cuttings used were those from the bases of vertically trained bines and bore two or more nodes in comparison with the layer cuttings which had only one node. Beard (1946) showed strap cuttings to be slightly superior to layer cuttings in the production of bedded sets).

It was apparent from visual inspection that the plants in this block were, in general, somewhat better than those elsewhere. This general appearance was reflected in the records of the weights of the sets produced which were heavier from strap cuttings than from layer cuttings. Survival of the plants of the variety OJ47 was also somewhat better than in the rest of the experiment, but OR55 did not show this effect. Both varieties gave rather more good cuttings per plant in this block than elsewhere.

Strap cuttings thus appeared to have slight but definite advantages over layer cuttings for use as parent plants in a layer bed of this type. It would therefore seem advisable to use them whenever possible, although normally the numbers available might be small.

SUBSIDIARY EXPERIMENT.

Concurrent with the experiment described above, a simplified trial of the same methods was made on a commercial farm. The same two varieties were used, planted in rather heavier soil than that at East Malling. The results from this trial are shown in Table III.

TABLE III.

Commercial trial of layering method.

Variety.	Parent material.	Layer length (ft.)	Spacing (ft.)	No. planted.	% Survival.	Good cuts per survivor.	Good cuts per plant planted.
OR55 ..	Sets	6	3	175	90	20·5	18·2
OJ47 ..	Sets	6	3	530	71	17·5	12·3
OR55 ..	Cuttings	2	1	160	84	11·3	9·6
		4	1	160	68	13·2	9·0
OJ47 ..	Cuttings	2	½	320	66	7·7	5·0
			1	160	78	8·1	6·3
			2	80	69	8·4	5·8
		4	½	320	56	11·2	6·3
			1	160	53	8·6	4·6
			2	80	55	9·8	5·4

Although the records from this trial could not be analysed they provide some indication of the results likely to be obtained under commercial conditions. These, in general, approximate fairly closely to those of the East Malling experiment. Somewhat better results were obtained with the sets of both varieties and with the cuttings of OR55 than in the comparable treatments at East Malling but the cuttings of OJ47 gave poorer results at wide spacing. The additional spacing treatment of 6 in. gave quite good results but the layering operation was somewhat impeded by tangling of the bines.

DISCUSSION.

The results of the experiments show that the bines from both cuttings and sets can be layered quite satisfactorily. Although sets give a somewhat larger yield of layer cuttings per plant, this is hardly sufficient to justify their use as parent plants in preference to cuttings.

Propagation from cuttings or sets has the considerable merit of eliminating the time lag inherent in all methods using mature plants only. This is likely to be of most value in the raising of new varieties or strains of hop which are in very short supply, as a ninefold annual increase will ensure a rapid building up of clonal stock. In breeding, for instance, a means of raising large numbers of plants from desirable parent seedlings should give considerable flexibility to the programme. The methods have also proved of value in propagating new varieties of hop bred or selected in the course of research on *Verticillium* Wilt.

It is apparent that even when layered, each plant grown from a cutting forms a good bedded set in addition to yielding layers. Thus, compared with the normal practice of planting cuttings for one year to produce bedded sets only, the layering process gives a considerable amount of *extra* propagating material. It seems likely that this reflects some extra growth of the aerial portions of the layered as compared with the unlayered bines. No data are yet available on this point but it was noted in the experiments that the layered bines appeared darker in colour than the unlayered ones and were apparently invigorated by the layering process. As the layers themselves form roots they presumably supply water and nutrients to the bines from the soil in which they are buried.

The methods employed might be used in commercial practice where rapid propagation was desired, when the spacing and layer lengths chosen would depend on the parent stock and land available. It is clear that very long layers should be avoided although some of the losses of plants from the long layer treatments could undoubtedly be eliminated by uncovering any buried bine tips after layering. If this were done it is likely that a layer length of 4 ft. for sets and 3 ft. for cuttings would prove satisfactory and probably give better yields than those obtained in the trials. There were few differences, except in weight of sets, between any of the spacing treatments and if economy in land, wirework, etc. were necessary, sets could be planted 2 ft. apart, and cuttings 1 ft. (or perhaps 6 in.) apart, in the rows. The rows could also probably be spaced at 4 ft. apart if suitable small cultivators were available. As an example of the potentialities of the methods it may be noted that from cuttings at 1 ft. spacing and with a layer length of 2 ft. the variety OR55 produced the equivalent of 78,000 cuttings per acre in the experiments.

One considerable disadvantage in methods using young parent plants is the impossibility of inspecting them adequately for the incidence of diseases such as

PLATE I.

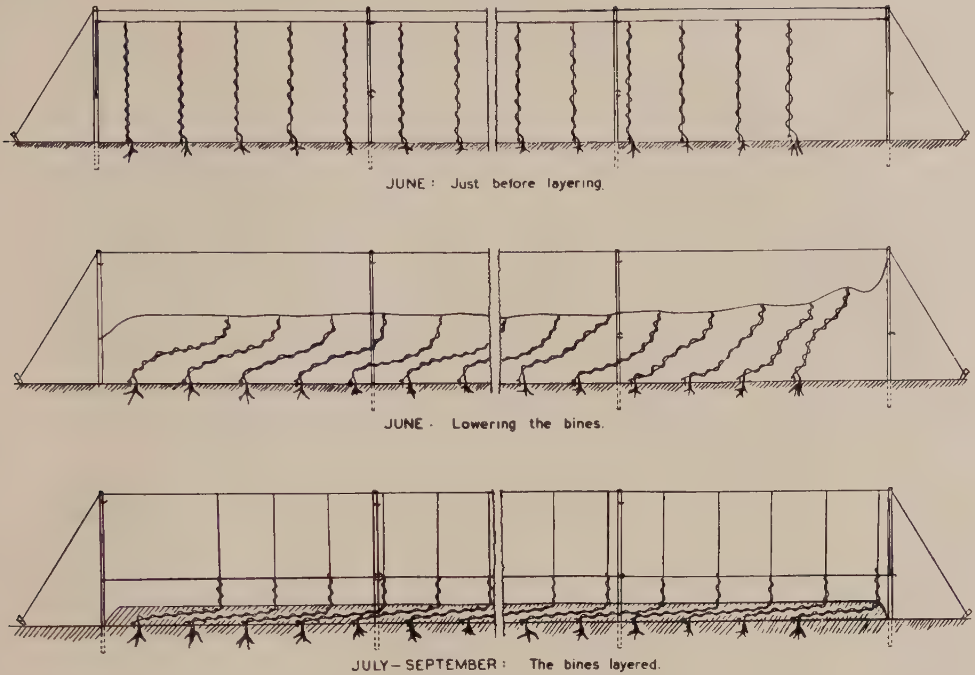
JUNE: Just before layering.
JUNE · Lowering the bines.
JULY–SEPTEMBER: The bines layered.

Fig. 1.
Diagram of layering process.

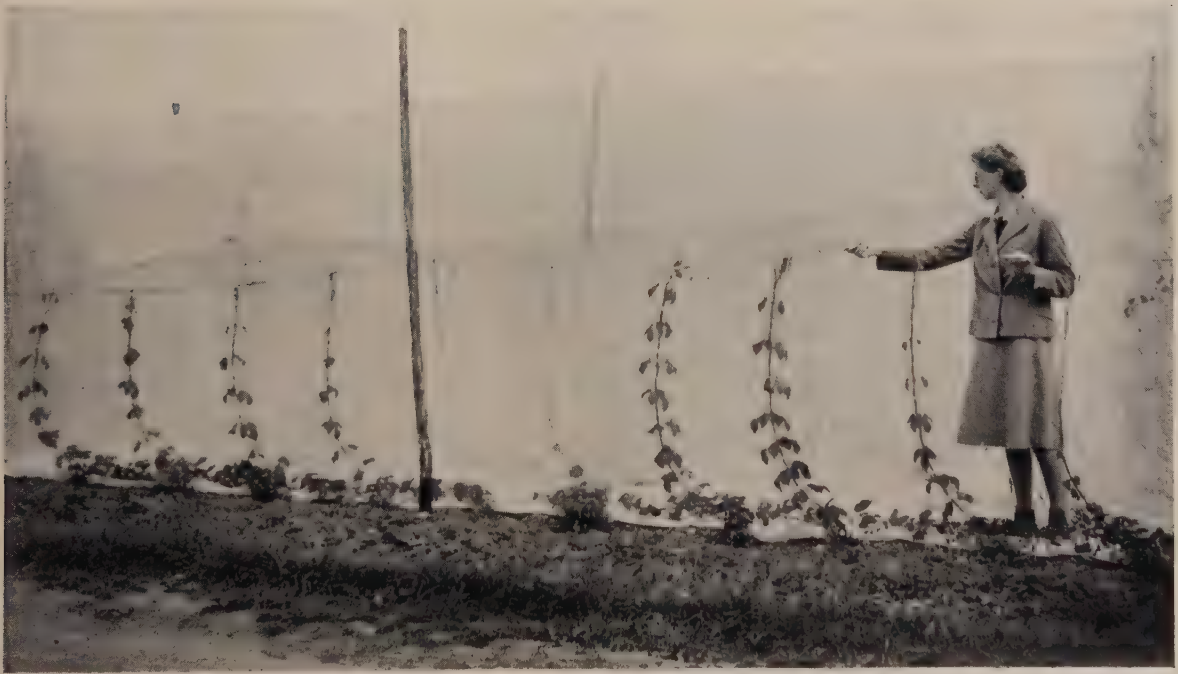

Fig. 2.
Lowering the bines. The plot illustrated is planted with bedded sets 3 ft. apart, the bines of which are being layered for 3 ft.

PLATE II.

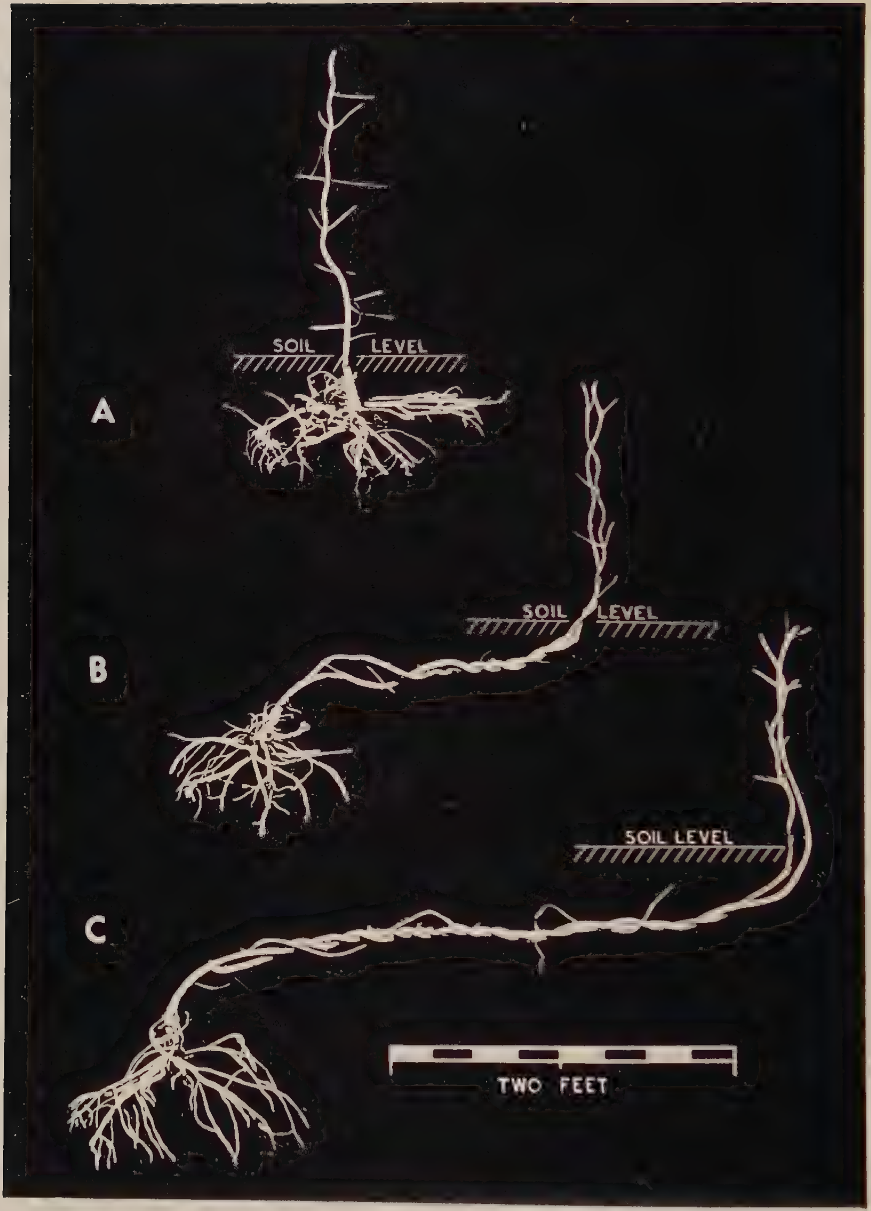
SOIL LEVEL
A
SOIL LEVEL
B
SOIL LEVEL
C
TWO FEET

FIG. 3.

Basal portions of plants (variety OR55) grown from cuttings in the experiments.

A unlayered.
B layered for two feet.
C layered for four feet.

Note that sets of plantable size were produced by all plants

Verticillium Wilt, Nettlehead or Mosaic. It would thus obviously be inadvisable to continue commercial propagation year after year from young plants except under conditions where the entry of such diseases was not possible. It is difficult at present to decide what these conditions are, as information on the means of dispersal of the diseases is small. It appears likely from present experience, however, that if care is taken to ensure the freedom of the parent stock from disease, isolation of a few miles from other hops should ensure the health of the progeny. It may, however, be advisable in commercial practice to establish on the propagation site a nuclear layer bed of permanent plants which can be inspected annually for disease. The cuttings from these plants would be grown and their bines layered. The new cuttings thus produced would not again be layered but would be grown into bedded sets for planting in hop fields, further cuttings for layering being obtained annually from the permanent layer bed after this had been certified free from disease.

ACKNOWLEDGMENTS.

The writers are indebted to the Statistical Section of the Research Station for advice on experimental layout and for analyses of the data. Thanks are also due to Miss J. Wickham and Miss H. Morgan for their work on the experiments and to Mr. R. F. Farrar for valued co-operation in the subsidiary experiment on a commercial farm.

SUMMARY.

An account is given of experiments on the layering of the bines on young hop plants grown from sets and cuttings. The effects of various planting and layering distances are described and it is shown that certain of the methods enable the stock to be multiplied from nine to fifteen times per annum. Cuttings which are layered also produce plantable bedded sets and the methods should prove of value in raising varieties or strains of hop in short supply.

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——— (1947b). Notes on varieties of hop resistant to Verticillium Wilt. Rep. E. Malling Res. Sta. for 1946 (1947), 157-9.

———, *Hitchcock, M. M.* and *Wilson, D. J.* (1948). Experiments on the layering of hops. I. The establishment of a permanent layer bed. J. hort. Sci., **24**, 200-205.

(*Received* 14/5/48.)

EFFECT OF HEAT PRETREATMENT ON THE EARLY FORCING OF SEAKALE

By I. F. STOREY and N. MONTGOMERY
Imperial College of Science and Technology, London, S.W.7

INTRODUCTION.

IN a study of the deterioration of certain commercial stocks of Seakale (*Crambe maritima* L.), Brown (1) showed that this effect was caused by the presence of an inferior type which tended to multiply unduly on account of its stronger rooting habit, the crop being carried on annually by means of root cuttings. The two types could be distinguished in the field from leaf characters and thereby the building up of a clean stock of the good forcing type could be effected.

Early forcings of the good type, begun for example in October, are always less satisfactory than those carried out later, say in December. The total forcing time is shorter in the latter case and the quality and quantity of the product is better. Thus in a December forcing it is usual to find that each crown shows 5-8 leaf stalks extended, and that these are arranged fairly compactly in a bunch, whereas a similar crown when forced early may have only 1-2 stalks fully extended with perhaps a few short ones in the centre. The outer stalks also tend to diverge and twist.

The following is an account of the experiments* started in the autumn of 1938 to determine whether the forcing of seakale crowns could be expedited by any of the standard treatments which have been developed for the breaking of bud dormancy. A comprehensive account of these treatments is given by Stiles (2).

EXPERIMENTAL.

The following treatments were applied to batches of 20 seakale crowns which had been lifted on 8th October, 1938. During the interval between lifting and the beginning of forcing (17th October), crowns which were not undergoing treatment were kept under moist soil in a cool shed.

Treatments.

1. Control.
2. Chilled for 2 days at 1°-3° C.
3. Chilled for 4 days at 1°-3° C.
4. Chilled for 6 days at 1°-3° C.
5. Heated in water for 3 hours at 30°-32° C.
6. Heated in water for 6 hours at 30°-32° C.
7. Heated in water for 9 hours at 30°-32° C.
8. Heated in water for 12 hours at 30°-32° C.
9. Exposed to ethylene chlorhydrin vapour for 24 hours.
10. Exposed to ethylene chlorhydrin vapour for 48 hours.
11. Exposed to concentrated sulphuric acid for 20 seconds.
12. Wounded by pricking.

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* The work was carried out over the period 1938-42 by I. F. S.; from 1944-7 by N. M.

Treatments 9-11 resulted in death of the buds; treatment 12 gave no stimulation to forcing and damaged the product. These methods were therefore abandoned. Table I gives the results obtained by the chilling and heating treatments.

TABLE I.

Results of chilling or heating Seakale crowns.

Treatment.	Wt. of Kale* (from 20 crowns).	Wt. of Crowns.	Kale/Crown ratio.
	gm.	gm.	
Control	308	730	0·42
Chilled 2 days	406	700	0·58
Chilled 4 days	406	760	0·53
Chilled 6 days	336	785	0·43
Heated 3 hours	406	780	0·52
Heated 6 hours	490	735	0·66
Heated 9 hours	378	780	0·48
Heated 12 hours	378	785	0·48

* As cut in the commercial manner, i.e. with about 1 in. of crown.

There was thus a clear indication that forcing was accelerated both by previous chilling and by heating of the crowns and that in both cases there was an optimum period of exposure. Facilities were not available for the elaboration of the former method, so that further experiments were directed to determining what was the optimum heat treatment. The results of two such experiments are shown in Text Figs. 1 and 2.

The most effective treatment was exposure to 35° C. for 3 hours, which gives sufficient latitude to make the treatment safe, although under experimental conditions equally good results might be obtained with a higher temperature operating for a shorter time.

Table II gives the records of four experiments carried out over the period 1939-42 in which pretreatment of the crowns at 35° C. for 3 hours was adopted as standard. The experimental material consisted of a number of clones of good forcing type, and in one case of a mixture of such. The data in the table show the percentage increase of kale weight, over that of the controls, obtained by the treatment.

In all of the 38 comparisons in Table II the heat treatment increased the yield, though by a percentage amount which varied considerably. The average overall increase is 37 per cent.

Plate I. Fig. 3, gives a typical example of the degree of acceleration in forcing obtained by preheating at 35° C. for 3 hours.

The results over the period 1944-47, with a mixture of clones of good forcing type taken from a commercial stock, are summarized in Table III.

An experiment was carried out in order to assess the effect of the hot water treatment in terms of the decrease in forcing time. Four replicates of 20 crowns each were cut after 4 weeks' forcing and the corresponding replicates after a further 3 weeks, with the results shown in Table IV.

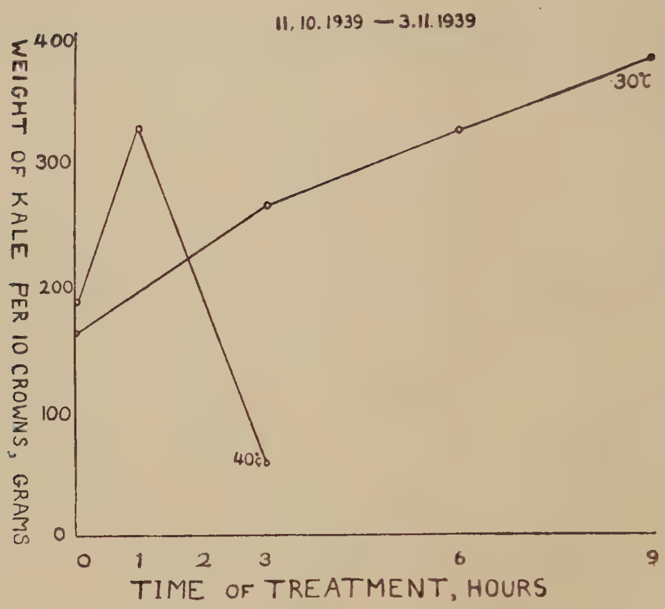
11.10.1939 — 3.11.1939
WEIGHT OF KALE PER 10 CROWNS, GRAMS
400
300
200
100
0
30°C
40°C
0
1
2
3
6
9
TIME OF TREATMENT, HOURS

FIG. 1.

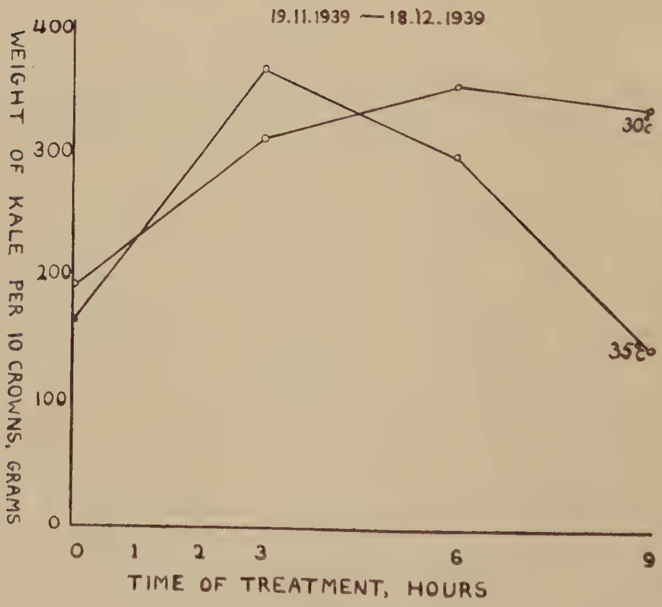
19.11.1939 — 18.12.1939
WEIGHT OF KALE PER 10 CROWNS, GRAMS
400
300
200
100
0
30°C
35°C
0
1
2
3
6
9
TIME OF TREATMENT, HOURS

FIG. 2.

TABLE II.

Pretreatment of Seakale crowns at 35° C. for 3 hours, 1939-42. Results expressed as percentage increase of Kale weight over that of the controls.

Clone	Exp. 1 22.12.38- 15.1.39.	Exp. 2 4.10.40- 24.10.40.	Exp. 3 15.11.41- 4.12.41.	Exp. 4 18.12.41- 12.1.42.
1*	—	22	39	—
2	24	56	—	—
3	50	23·5	—	—
4	—	—	66	—
5	—	11	23	—
6	22	50	53	—
7	9	26	—	27
8	—	—	70	—
9	6	—	44·5	—
10	32·5	83	44·5	—
11	44	64	—	22
12	—	—	—	10
13	62	50	19·5	—
14	—	—	35·5	—
15	14·5	17	100	—
16	20·5	4	49	—
17	38·5	49·5	43	—

* No. 1. consisted of a mixture of good clones.

TABLE III.

Pretreatment of Seakale crowns at 35° C. for 3 hours, 1944-47.

	Dec. 1944.	Nov. 1945.	Oct. 1947.	Dec. 1947.
Total No. of Crowns . .	640	320	256	352
Treatment Mean* . .	137	134	133	164
Control Mean*	118	93	114	128
t	4·75	7·01	2·20	5·17
n	38	14	14	20
P	$<\cdot01$	$<\cdot01$	$<\cdot05$	$<\cdot01$

* Grams of Kale per 100 grams of crown weight.

TABLE IV.

Kale cut from pretreated crowns forced for 4 weeks and 7 weeks. Results expressed as grams Kale per 100 grams crown wt.

Batch.	After 4 weeks.		After 7 weeks.	
	Control.	Treatment.	Control.	Treatment.
A.	96	147	167	190
B.	87	147	146	178
C.	110	149	170	242
D.	94	116	133	178
Average	97	140	153	197

After 7 weeks' forcing the controls had slightly exceeded the yields obtained from the treated crowns after 4 weeks. The saving in time thus amounts to 2-3 weeks over a period of 6 weeks, i.e. it may be estimated conservatively as about one-third of the forcing period.

The general result therefore is that, by preheating the crowns in water at 35° C. for 3 hours, the weight of kale produced in a given time is increased by about one-third, or alternatively that the same weight is produced in one-third less time. More recently, however, it has appeared that certain new types raised from seed give consistently higher yields than does the commercial mixture, especially in early forcings. Table V records the behaviour of one of these new types, III 37, compared with that of commercial mixture in forcings at two dates.

TABLE V.

Comparison of the results of forcing Seakale, (a) of the new seedling III 37 and (b) of the commercial mixture.

	Oct. 1947.	Dec. 1947.
Total No. of leaves	80	80
Type III 37, mean wt. of kale*	234	270
Mixture, mean wt. of kale*	131	143
t	12	12·1
n	6	6
P	<·01	<·01

* Grams Kale per 100 grams crown wt.

In the same experiments preheating at 35° C. for 3 hours increased the yield of commercial mixture by 17 per cent. and 28 per cent. in October and December respectively, the corresponding increases shown by Type III 37 over commercial mixture were 78 per cent. and 89 per cent. The behaviour of certain selected new types will be described in a later paper, but the results quoted suggest that, for early forcings, it would be preferable to use such a type as III 37 rather than to apply the warm water treatment to the commercial type.

SUMMARY.

By preheating seakale crowns in water at 35° C. for 3 hours the yield of kale is increased by approximately one-third, or alternatively the duration of the forcing period is reduced by a like amount. Much greater increases in yield are obtained by use of new early-forcing types of seakale.

This work arose from a study of seakale clones made by Professor W. Brown, F.R.S., and the authors wish to express their thanks to him for his advice and assistance.

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(1) *Brown, W.* (1937). A study of the deterioration of seakale stocks, with notes on some diseases of that crop. J. Pomol., **15**, 69.
(2) *Stiles, W.* (1936). An introduction to the principles of Plant Physiology.

(*Received* 7/6/48.)

PLATE I.

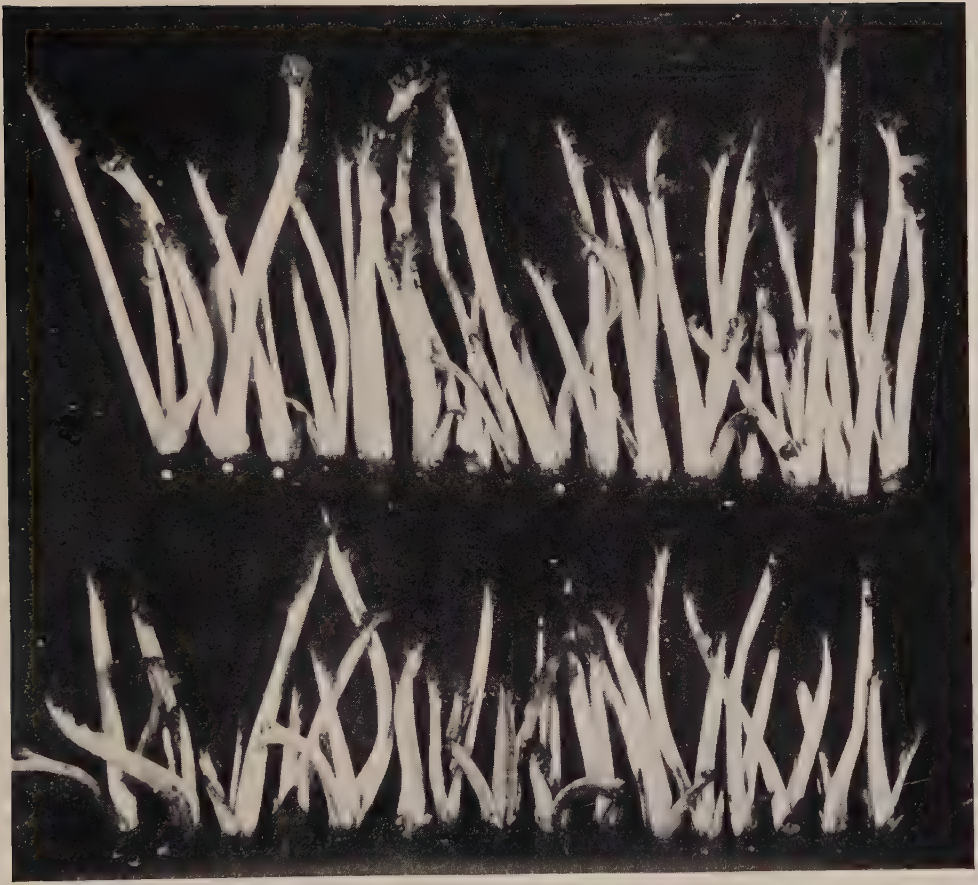

FIG. 3.

A typical example of the degree of acceleration in forcing obtained by preheating Seakale at 35°C. for 3 hours.

VERTICILLIUM WILT OF THE HOP *(HUMULUS LUPULUS)*

V. THE INFLUENCE OF NUTRITION ON THE REACTION OF THE HOP PLANT TO INFECTION WITH *VERTICILLIUM ALBO-ATRUM*

By **W. G. KEYWORTH**
East Malling Research Station, Kent
and
E. J. HEWITT
Long Ashton Research Station, Bristol

THE effects of the nutrition of plants on their reaction to disease have been studied by many workers. Much of the literature relating to the general aspects of this work has been reviewed by Wingard (1941) and that dealing with diseases caused by root fungi by Garrett (1944). Of the work on vascular diseases, the following may be noted.

Spencer and McNew (1938) reported that the wilt disease of sweet corn caused by *Phytomonas stewarti* was increased in severity by increase in the nitrogen or phosphorus supply to the plants ; the disease was also more severe when the potassium supply was either in excess or deficient. On the other hand, seedlings dwarfed by reductions in nitrogen and phosphorus were only slightly affected. McNew and Spencer (1939) attributed the effect of nitrogen in increasing the severity of the disease to direct action on the growth of the pathogen in the vascular tissue.

V. H. Young (1938), Dick and Tisdale (1938) and Smith (1940) noted a reduction in the severity of cotton wilt caused by *Fusarium vasinfectum* when the crop was liberally manured with potash fertilizers. This effect was confirmed by P. A. Young (1943). Cook (1937), working on the wilt disease of tomatoes caused by *F. lycopersici*, found a positive correlation between the supply of nitrogen and the severity of the symptoms. Stoddard (1947) demonstrated a similar effect in respect of the wilt disease of muskmelons caused by *F. bulbigenum var. niveum*.

Roberts (1943, 1944), working with tomatoes, reported that the effect of the wilt disease caused by *Verticillium albo-atrum* was increased by the application of nitrogenous manures. She noted that reduction in nitrogen supply both reduced the numbers of infected plants and retarded the development of the pathogen in such plants.

The present sand-culture pot experiments were started in 1944 in conjunction with the general studies on *Verticillium* wilt of the hop in progress at the East Malling Research Station, and were designed to be complementary to field studies on the variation in disease intensity which is a feature of this disease in commercial plantations.

Two experiments were carried out, the first during 1944-45 and the second in 1946-47.

EXPERIMENT NO. 1 (1944-45).

(*a*) *1944.* 135 hop cuttings (var. Fuggle, East Malling N clone) were planted on 24th March, 1944 in 12 in. waxed flower pots in the open air and the pots were sheltered by wooden boxes.

The plants were divided into 9 sets of 15 plants ; one set was planted in 9 kilo. greenhouse compost and the others in 14 kilo. silica-sand. The treatments applied to these latter were modifications of a complete nutrient solution of the following composition* :—

Nutrient salts.	*Gm. wt. in 10 litres.*
KNO_3	2·1
$CaNO_3$ (anhyd.)	6·6
$NaNO_3$	8·7 (after 2nd August, 1944)
$NaH_2PO_4 \cdot 2H_2O$	2·0
$MgSO_4 \cdot 7H_2O$	4·0
Ferric citrate	0·195
$MnSO_4 \cdot 4H_2O$	0·0121
$ZnSO_4 \cdot 7H_2O$	0·0022
$CuSO_4 \cdot 5H_2O$	0·002
H_3BO_3	0·0186
$(NH_4)_4Mo_7O_{24} \cdot 4H_2O$	0·00028

One set of 15 plants received this solution. The other treatments were modifications of this solution containing respectively twice normal nitrogen (as extra $NaNO_3$), twice normal phosphorus (as extra $NaH_2PO_4 \cdot 2H_2O$), twice normal potassium (as extra K_2SO_4), one-third normal nitrogen, one-quarter normal phosphorus, no potassium, no calcium, and no magnesium.

The plants were grown during 1944 without inoculation to allow them to develop the desired nutrient status. During this season, deficiency symptoms developed on the plants supplied with solutions lacking certain elements. Descriptions of these symptoms have been published already by Hewitt (1945, 1946). They confirmed and extended earlier observations made by Beard (1932). Tissue tests were made on petioles collected at the 5-6 ft. level (Nicholas and Jones (1945)). The visual observations and tissue tests (see Table I) showed that all the plants, with the exception of those receiving twice normal nitrogen, were somewhat low in this element and that many of the plants contained only medium or less than medium amounts of phosphorus at the first sampling date. The nitrogen level was doubled throughout on 2nd August to raise the nitrogen status generally, except in the low N and the compost series which were left unchanged. The stems were cut down in October and the plants kept in the greenhouse until the next spring.

(*b*) *1945.* The plants were put in the open again in mid-March. As the compost had a pH of 5·96 and the 1944 tissue tests had indicated a low calcium status, lime was added to it in March, 1945, at the rate of 4 gm. per pot to represent 10 cwt. per acre.

Nine plants in each set were inoculated on 6th April, 1945, by adding 5 gms. of chopped hop stem infected with a progressive strain of *Verticillium albo-atrum* (Keyworth (1942), Isaac and Keyworth (1948)), to each pot, this being distributed in three holes in the sand or compost at about 3 in. from the plant. In order to test

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* Further details of the methods used have already been published—Hewitt (1944, 1945).

TABLE I.

Estimates of nutrients present in petioles, made by the tissue test method. Seasons 1944-45.

1944 Test for	Treatments (Sampled 2.8.44).									
	Compost.	Complete.	$\frac{1}{3}$ N.	$\frac{1}{4}$ P.	−K.	−Ca.	−Mg.	2N.	2P.	2K.
Ca	L	M	L+	M+	M−	L	M−	M−	H+	M+
K	H−	M−	H−	M	L	M−	M+	M	M+	H−
Mg	M	M	M−	M	M	M−	L	M+	M	M
PO_4	L+	M	M	L	M−	M−	M+	M−	H−	L+

1945 Test for	Treatments (Sampled 29.6.45).									
	Compost.†	Complete.*	$\frac{1}{6}$ N*	$\frac{1}{4}$ P.	−K.	−Ca.	−Mg.	2N*	2P.	2K.
Ca	H	M	M+	M	M	L+	M	H−	M	M
K	H−	L+	H−	M	L	M−	M+	L	M−	M+
Mg	M−	H−	L+	M−	H−	M	M−	H	M+	M
Mn	H++	Tr+	H	Tr+	Tr	Tr	Tr	Tr+	Tr+	M
PO_4	H	M+	H−	L	H−	M	H	M+	H	M−
NO_3	M	M	M	M	M	M	M	H−	M	M

L = Low. M = Medium. H = High. Tr = Trace.

* The nitrogen level in the complete and 2N series was doubled on 2.8.44 leaving Low N as $\frac{1}{6}$N in 1945.

† The compost was limed in March, 1945.

the nutrient effect of the hop stem, 5 gms. of sterilized stem was added to each of three of the six control plants in each set, the other three being untreated. No effect was seen.

Wilt records were made from mid-June onwards when the first symptoms were seen—on one plant treated with complete nutrient and two with the high potassium solution. Tissue tests were again made (on the control plants) at this time (Table I). Deficiency symptoms were also apparent, and were particularly marked in the low nitrogen, potassium, calcium and magnesium treatments

A final symptom record was made on 5th September, 1945. The wood of all the stems was examined at a height of three inches from the base for the brown discoloration indicative of the presence of *Verticillium*. If this was not seen, isolation tests for *Verticillium* were made of the wood at the base of the stems and in the crowns of the plants, but it was not practicable to test the roots in this way. Results are given in Table II.

This record indicates that reduction of any of the elements nitrogen, phosphorus or potassium resulted in the reduction of both disease incidence and severity as measured by the visible symptoms. In each case some plants were found to be infected, although not showing symptoms, and there were also others from which the pathogen could not be isolated. The elimination of magnesium had a similar but less marked effect on the expression of wilt symptoms but *Verticillium* was isolated from all the inoculated plants in this treatment. The elimination of calcium or the supply of additional nitrogen, phosphorus or potassium did not apparently affect wilt incidence or severity.

TABLE II.

Record made on 5.9.45 of the inoculated plants receiving the various nutrients. The record includes visible symptoms, presence of brown wood and occurrence of Verticillium *in the stems or crowns of apparently healthy plants.*

Treatment.	Symptom record.					Other plants from which Verticillium was isolated.	Total infected.	Av. wt. stems on control plants.
	D	W+	W	MW	Brown wood only.			
Complete nutrient ..	2	1	4	1	1	–	9	363 gm.
2N	3	1	5	–	–	–	9	218 gm.
2P	2	1	3	2	1	–	9	333 gm.
2K	3	2	3	–	1	–	9	360 gm.
No Ca	1	2	4	2	–	–	9	99 gm.
No Mg	–	–	5	–	–	3	8	66 gm.
No K	–	–	2	–	–	4	6	170 gm.
⅙N	1	–	–	–	1	4	6	167 gm.
¼P	–	–	1	–	2	3	6	207 gm.
Greenhouse compost ..	–	–	–	–	–	–	–	50 gm.

Note.—The external symptoms are classified as follows in this table and in Tables III and V.
D = whole plant dead.
W+ = Severe Wilt—most of the stems dead or badly wilted.
W = Wilt—some dead or wilted stems—remaining stems apparently normal.
MW = Mild Wilt—mild symptoms on some or all of the stems.

As will be seen, the plants growing in greenhouse compost showed no infection of the crowns or stem bases. The growth of the plants was very poor and the 1945 tissue tests indicated that they were deficient in nitrogen. These plants and those in the low nitrogen series, also had a high manganese status (3 and 2 p.p.m. respectively).

EXPERIMENT NO. 2 (1946-47).

The second experiment was started in 1946. The basic solution was similar to that used in 1944-45, the modification for each treatment being as follows :

1. Complete nutrient (calcium at 200 p.p.m.)
2. High calcium—400 p.p.m. (replacing sodium).
3. High potassium—160 p.p.m.
4. High phosphorus—80 p.p.m.
5. High manganese—6 p.p.m.
6. Low nitrogen—140 p.p.m.
7. Low potassium—20 p.p.m.
8. Low phosphorus—20 p.p.m.
9. Compost A—Soil 2 parts, sand 1 part, peat 1 part, *plus* (per bushel)—6 oz. hoof and horn, 3 oz. superphosphate, 1½ oz. sulphate of potash and ¾ oz. ground lime.
10. Compost B—As Compost A, but 1¼ oz. ammonium nitrate per bushel substituted for hoof and horn.

The treatments given in this second experiment were selected in order to take into account the indications shown in the 1944-45 experiment and also other evidence from field experiments by Dr. Pizer and his colleagues of Wye College (in the course of publication). These showed that the occurrence of severe outbreaks of Wilt in commercial plantations might be correlated with the calcium content of the plants. As well as the high calcium treatment shown, therefore, a number of low calcium treatments were included, but the results from these were invalidated by the fact that the waxed pots used were able to supply appreciable amounts of calcium (Hewitt 1946) and the tissue tests made on the plants in these series showed that their calcium status did not reflect the treatments given.

As a notable feature of the results obtained in 1944-45 was the freedom from Wilt of the plants in the greenhouse compost series, two composts were used in this new experiment,* nitrogen being supplied in one in an organic, and in the other in an inorganic, form. Both composts were limed to approximately pH 6·5. As already noted, two of the sets of plants with least Wilt in the previous experiment showed a high manganese status and a high manganese treatment was therefore included in the new series to determine its effect on the disease.

1946. Eight plants in each series were inoculated on 5th May. In an attempt to increase the sensitivity of the experiment, only 2 gms. of infected hop stem was added to each pot. Symptoms were recorded as for the 1944-45 experiment, a final record being made on 13th September.

TABLE III.

Record made on 13.9.46 of the plants inoculated in May, 1946, showing visible symptoms and the occurrence of Verticillium *in the stems of apparently healthy plants.*

Treatments.	No. of plants inoculated.	Symptom Record.					
		D	W+	W	MW	Internal stem infection only.	Total infected.
Complete nutrient . .	8	1	3	3	—	1	8
High Ca	8	2	1	1	1	2	7
High K	7	—	—	—	3	3	6
High P	7	1	1	1	2	1	6
High Mn	8	1	3	—	1	2	7
Low N	8	—	1	—	—	2	3
Low K	7	4	—	—	—	3	7
Low P	6	—	1	2	1	1	5
Compost A	7	—	4	1	—	1	6
Compost B	7	7	—	—	—	—	7

---

* *Note.*—It had originally been intended to use an organic form of phosphorus (6 oz. steamed bone flour per bushel) in Compost A, but most of the cuttings planted in a compost thus prepared were attacked and killed by an unidentified soil fungus. Tissue tests and visual observations showed the plants in this compost to be deficient in phosphorus. Thus the compost (A) shown above was prepared and new cuttings planted, which grew satisfactorily and had a normal phosphate status.

It is apparent from this record that the low nitrogen treatment again reduced disease incidence and severity. The other results do not correspond so closely with those obtained in the previous experiment. There was a less definite reduction in disease severity in the plants receiving reduced phosphorus or potassium, but a noticeable reduction in those receiving high potassium. The disease was severe in the plants growing in both composts and in both the growth of the control plants was satisfactory. The high manganese treatment had no marked effect on symptoms.

Tissue tests were made of the control plants (Table IV).

TABLE IV.

Estimates of nutrients present in petioles, made by the tissue test method. Seasons 1946-47.

1946 Test for	Treatments (sampled 29.7.46).									
	Compost A.	Compost B.	Complete.	Low N.	Low P.	Low K.	High P.	High K.	High Ca.	High Mn.
Ca	H	M+	H−	M+	M	H−	M+	H−	M+	H−
K	H−	H−	H−	H	H	L+	M+	H	M	H−
Mg	H	H−	M−	L+	M−	M−	M−	L+	M−	M−
Mn	Tr+	—	—	Tr	—	—	—	—	—	H+
PO_4	H	H	L	M	L−	M−	M+	L+	M−	L+
NO_3	H	H+	*Not detected in any of these treatments.*						L.	N.D.

1947 Test for	Treatments (sampled 20.6.47).									
	Compost A.	Compost B.	Complete.	Low N.	Low P.	Low K.	High P.	High K.	High Ca.	High Mn.
Ca	H−	H−	M	M+	M+	M+	M	M	H−	H−
K	H−	H−	H−	M+	H−	M+	H−	H−	M−	H−
Mg	M	L+	M+	M	M	M	M−	M	M	M−
Mn	H+	—	—	—	—	Tr	—	Tr	Tr	H+
PO_4	H+	H++	M+	M+	L	L	M+	L	L+	H−

1947. During the winter of 1946-47 a number of the inoculated and control plants died. Therefore, although it was intended to inoculate a further 8 plants in each series in 1947, in some cases the numbers available permitted only 6 or 7 to be inoculated. This was done on the 17th April, using the same methods as in 1946. As their pot identity numbers had been lost, the plants receiving high and low potassium and phosphorus could not be inoculated and these series were discarded.

Symptoms were recorded as in previous years. The final record made on 18th September on the plants inoculated in 1946 and 1947 is given in Table V.

Taking into account the mortality to be expected among the plants showing severe wilt in 1946, the results from the inoculations in both years correspond fairly closely. The most outstanding result is the small number of plants from which *Verticillium* was isolated and the reduced severity of symptoms, in the low nitrogen treatment.

Table V.

Record made on 18.9.47 of the plants inoculated in May, 1946 and April, 1947. The record includes visible symptoms and occurrence of Verticillium *in the stems of apparently healthy plants.*

Treatment.	No. of plants inoculated. a. 1946. b. 1947.	Symptom Record.					
		D	W+	W	MW	Internal infection only.	Total infected.
Complete nutrient	a. 8	4	1	1	1	1	16
	b. 8	2	6	–	–	–	
High Ca	a. 7	3	–	–	1	3	14
	b. 8	–	1	1	2	3	
High Mn	a. 8	3	1	3	–	–	15
	b. 8	–	4	4	–	–	
Low N	a. 8	1	–	–	1	1	5
	b. 8	–	–	1	1	–	
Compost A	a. 7	3	–	1	–	1	13
	b. 8	5	3	–	–	–	
Compost B	a. 7	7	–	–	–	–	10
	b. 6	3	–	–	–	–	

DISCUSSION.

Of the treatments tested in the two experiments, a reduction in the nitrogen supply of the plants caused the most marked and consistent reduction in disease incidence and severity. This result agrees closely with those obtained by some of the other workers already quoted—notably Roberts (1943).

Many of the nitrogen deficient plants from which *Verticillium* was isolated did not show any external symptoms and the fungus failed to grow farther than the crowns of the plants or lower parts of the stems. It may thus be concluded that the conditions prevailing in these plants inhibited both the growth of the pathogen and its ability to cause wilting of the plants. Owing to the difficulty of a critical examination of the root systems, it was not possible to determine whether the fungus was present in the roots of the other, " apparently healthy " inoculated plants, but it seems likely that this was so and that its growth was even more strongly inhibited than in those plants in which it was found in the rootstocks and stems. No information is available on the host-parasite relationship in the nitrogen deficient plants, but two possible reasons for the effects observed are enhanced plant resistance or a reduction in the vigour or wilting power of the fungus, caused by the reduced nutrient supply in the vascular tracts.

McNew and Spencer (1939) suggested that the reduction in growth of *Phytomonas stewarti* in nitrogen deficient maize plants was due to the starvation of the pathogen by the depletion of available nitrogen in the tracheal systems of the plants. In this connection it should be noted, however, that in neither of the present experiments was there any correlation between the severity of wilt symptoms and the nitrate content of the tissue extracts of the petioles. With the exception of the high N treatment, all the plants in the 1944-45 experiment, including those in the low N treatment, contained a medium amount of nitrate. In the 1946-47 experiment, only the plants grown in compost contained appreciable quantities of nitrate.

Certain other treatments caused a reduction in disease incidence and severity, viz., reductions in potassium, phosphorus and, to a lesser extent, magnesium, in the 1944-45 experiment. In this experiment also, those plants growing in compost showed no disease symptoms. It is significant that all these treatments resulted in some degree of plant starvation and it is thus conceivable that the resultant reduction in disease may be related more to general plant starvation than to lack of any particular element. The evidence on this is inconclusive, however, as the effects observed were not again seen in the 1946-47 experiment.

One disadvantage of the inoculation method used should be noted. As the inoculum was put in the sand and not directly into the plants themselves, fungal growth in the different treatments may have been influenced by the nutrient content of the sand before invasion of the plants occurred. However, owing to the abundant root growth of the plants which ensured contact between roots and inoculum, and owing to the fact that plants known to be infected in 1946 continued to show consistent symptoms in 1947, it is not thought likely that the results obtained were affected by any variation in growth of the inocula in the different treatments prior to plant invasion.

SUMMARY.

A description is given of two experiments extending over four years on the effect of *Verticillium albo-atrum* on hops growing in sand cultures supplied with various nutrient solutions. The plants were grown under conditions involving both deficiencies and excesses of most of the major elements necessary for plant growth.

In both experiments nitrogen-deficient plants showed much less severe symptoms than those grown in complete nutrient. In the first experiment only, deficiencies of phosphorus and potassium (and possibly of magnesium) produced the same effect. Other modifications in the nutrients supplied did not result in any marked alteration in the final symptom status of the plants.

ACKNOWLEDGMENTS.

The authors wish to express their thanks to Professor T. Wallace, at whose suggestion these experiments were made, for his help and advice throughout. Thanks are due also to Dr. E. H. Wilkinson and Miss M. M. Hitchcock for their help in the inoculation and symptom recording, and to Dr. D. J. D. Nicholas for assistance with the chemical tissue tests in 1944 and 1945.

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(*Received* 27/8/48.)

COVER CROPS FOR FRUIT PLANTATIONS

IV. LONG-TERM LEYS AND PERMANENT SWARDS

By W. S. ROGERS, TH. RAPTOPOULOS and D. W. P. GREENHAM
East Malling Research Station, Kent

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INTRODUCTION.

The oldest form of orchard cover cropping in England is undoubtedly the traditional grass orchard, grazed by animals, and with tall standard trees, as described by Hoare (1928). This tree form is not well adapted to the pruning, spraying and careful picking which are needed to obtain high quality apples and pears, and now survives mainly in West Country cider orchards ; it is also generally used in cherry orchards in Kent and elsewhere.

The more recent alternative to the home orchard and " the old apple tree " has been the modern plantation of bush-form apples, on standardized rootstocks of moderate vigour, kept clean cultivated for most of the year, and heavily manured with organic and other manures. As organic manures have become scarcer, however, it has become apparent that continuous clean cultivation of orchard soils, *without* the addition of organic manures, has resulted in progressive lessening of the soil organic matter, and deterioration of soil structure. This in turn has also sometimes led to soil erosion, and various deficiency symptoms have been of frequent occurrence in the trees. There is evidence that the addition of organic matter, by grassing down or by other forms of cover cropping, has led to an amelioration of these conditions (Wallace, 1928 ; Collison and Carleton, 1942 ; Rogers and Raptopoulos, 1945).

Much work on orchard cover cropping has been done in the U.S.A. and elsewhere, and the relevant literature was reviewed in the first paper of this series (Rogers and Raptopoulos, 1945). A series of studies on various types of orchard cover cropping was started at East Malling in 1940, and accounts of the use of short-term leys and annual cover crops have already been published (Rogers and Raptopoulos, 1945, 1946).

No definition has been laid down for the duration of a " long-term ley " in the present study, but it was assumed at the outset that, just as it has been found advantageous on the arable farm to plough up old pastures at intervals and re-seed them either at once or after a cycle of arable crops, so it might be useful to plough up grass swards in the orchards and replace them later with other swards. This practice, in old and neglected grass orchards, has proved successful during the recent war years but little or no experimental evidence is so far available.

The present study is of the effect of swards, sown in 1940, on the soil and on the trees, up to the crop of 1946. Some of the swards were turned in in 1943 and some have remained up to the time of writing and may now be left for a much longer period. It is therefore appropriate to consider the study as one of both long-term leys and permanent swards. It is obvious that many years must elapse before final conclusions on the long-term effects can be drawn ; but the results obtained so far have provided some interesting contrasts and have given some indication of the principles that should be followed in sward culture in fruit plantations under English conditions.

It is unfortunate that the material available did not permit the choice of less variable trees, and better replication and randomization of the plots. But even the present relatively simple design has provided a large amount of interesting new data. It will be seen that calculation of the statistical significance of differences is possible in some but not all cases and statistical analyses have been made wherever it seemed desirable and was possible.

MATERIAL AND METHODS.

SOIL AND ASPECT.

The experiment occupies a 3½ acre plantation sloping gently down towards the north. The soil is a medium sandy loam, overlying ragstone, and is partly of the Malling Series and partly of the allied Barming Series (Bane and Gethin-Jones, 1934). The soil of this plantation is rather shallower than in many parts of the Research Station and although the rock lies at a depth of 3 ft. or more in some parts of the plantation, in others it occurs at 2 ft. and, in one place, at 18 in. Samples taken near the beginning of this trial (28th June, 1940) were analysed at Wye College by Dr. N. H. Pizer. In general the pH was found to be about 8, the lime requirement was *nil*, and the soil was rich in both available phosphorus and available potassium. Organic matter content was determined both on these 1940 samples and on samples taken in 1943; nitrogen content was determined in 1943 (see p. 243).

RAINFALL.

The average annual rainfall at East Malling for the period 1915-1944 inclusive was 26·24 in. The actual totals for the years 1940-46 were 27·01 in., 26·00 in., 26·25 in., 25·70 in., 25·40 in., 22·48 in. and 32·43 in.

TREES.

The plantation used for this investigation was part of a 30-year-old commercial apple orchard of bush Worcester Pearmain (see Fig. 1). These trees were not ideal for experimental purposes, being rather variable; some were on rootstock M.II, some on seedling stocks, and most of them were not in a vigorous state. There were some bush Cox's Orange Pippin and standard Bramley's Seedling in guard rows.

TREATMENTS.

The trial was, to a certain extent, a "pilot" experiment in which the treatments were developed and changed as the experiment proceeded, for remarkably little precise information on the effect of swards on fruit trees has hitherto been available. Indeed this appears to be the first such experiment in England, in which replicated plots have been used.

The layout of the trial is shown in Fig. 2. The main comparisons were two grass and clover mixtures, A and B, compared with each other and with a clean cultivated strip, two frequencies of cutting (up till 1943), and five manurial treatments. A third grass mixture, C (fine grasses), occupied an adjacent triangle, as shown.

With the two main grass mixtures there were 180 experimental trees, in three randomized blocks arranged so that the cutting treatments ran right through the strip for ease of management, with guard rows between all treatments. The main plots were divided into small sub-plots for the different manurial treatments, each of which occupied an area of 192 square yards and contained three experimental trees, with guard trees all round. The extra plot under mixture C contained seven similar sub-plots. The single clean cultivated strip contained 30 experimental trees. The distance between trees was 17 ft.

Seeds mixtures. The composition of the seeds mixtures is shown in Table I.

PLATE I.

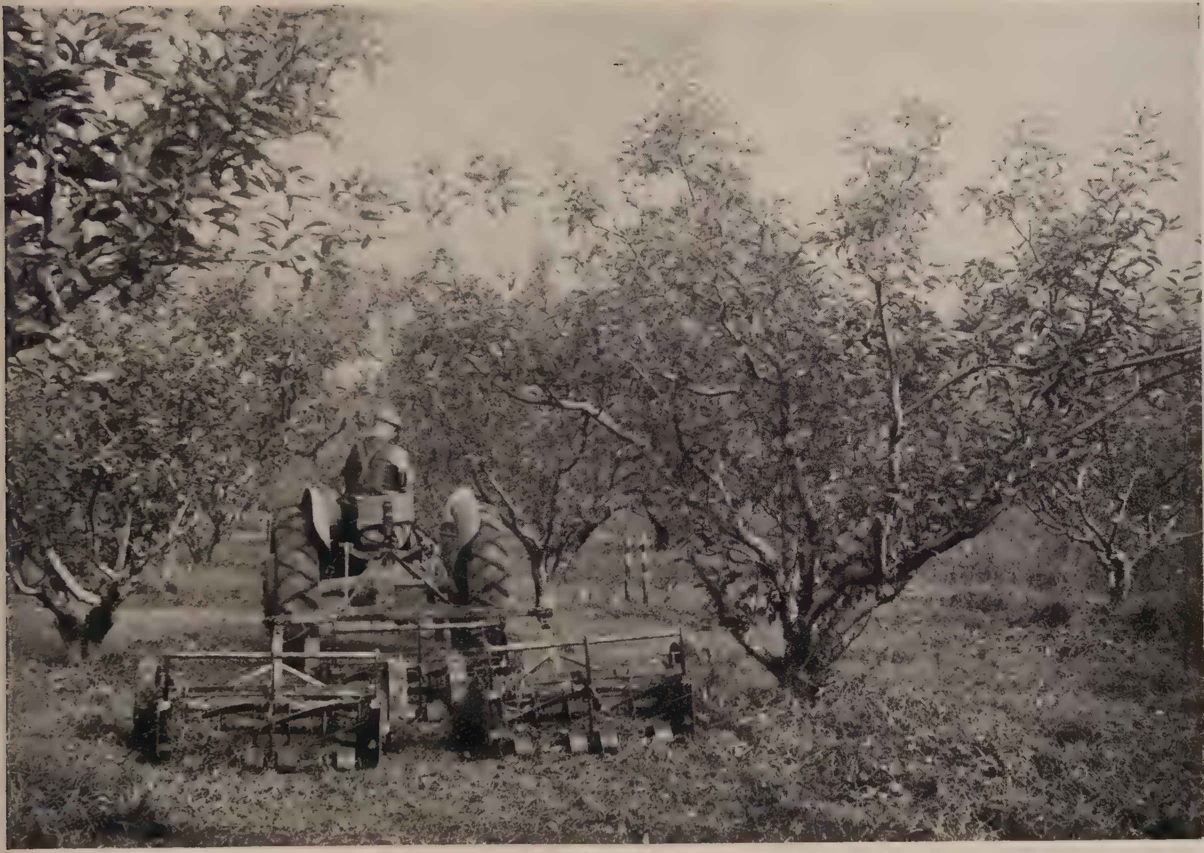
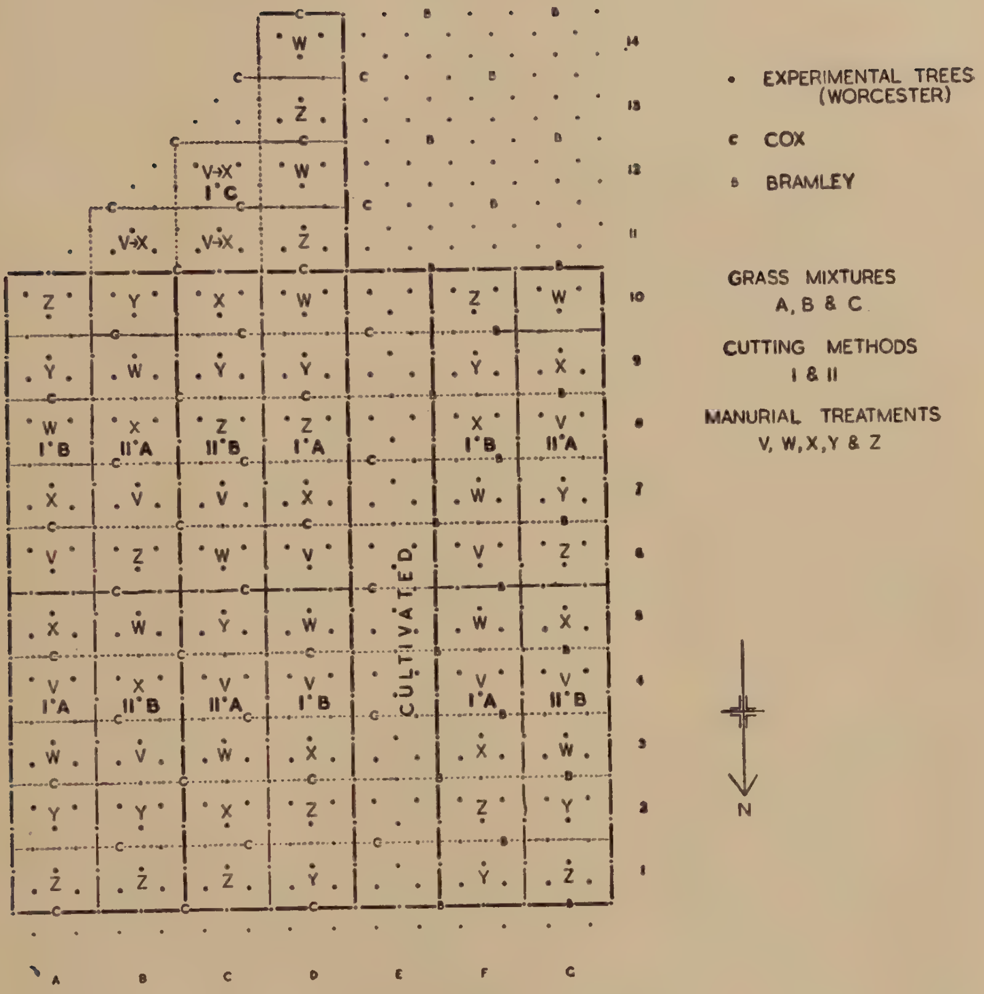


FIG. 1.
Gangmower cutting grass in the experimental orchard.



• EXPERIMENTAL TREES (WORCESTER)
c COX
B BRAMLEY
GRASS MIXTURES A, B & C
CUTTING METHODS I & II
MANURIAL TREATMENTS V, W, X, Y & Z
CULTIVATED
N
A
B
C
D
E
F
G
1
2
3
4
5
6
7
8
9
10
11
12
13
14

Fig. 2.

Plan of experimental plantation.

TABLE I.

Composition of seeds mixtures.

	lb./acre.		
	A.	B.	C.
Perennial ryegrass (*Lolium perenne*) (commercial)	8	20	–
Perennial ryegrass (*Lolium perenne*) (S. 23)	6	–	6
Cocksfoot (*Dactylis glomerata*) (S. 143) ..	10	–	–
Rough stalked meadow grass (*Poa trivialis*)	2	4	4
Wild white clover (*Trifolium repens* var. *sylvestre*)	2	1	1
Timothy (*Phleum pratense*)	–	4	–
Late red clover (*Trifolium pratense* var. *serotinum*)	–	3	–
Broad red clover (*Trifolium pratense* var. *praecox*)	–	2	–
Browntop (*Agrostis tenuis*)	–	–	2
Sheep's fescue (*Festuca ovina*)	–	–	2

It can be seen that the main differences between mixtures A and B are that B contained 5 lb. of red clover besides 1 lb. of wild white, while A contained only 2 lb. of wild white clover. A also had 10 lb. of cocksfoot while B had none, and A had 6 of its 14 lb. of ryegrass of the Aberystwyth S. 23 strain, while B had only commercial perennial ryegrass. Mixture C was intended to give a sward mainly of fine lawn grasses but with relatively low seeding rates to keep down the cost. It was hoped that such a mixture, giving less vigorous growth, would also lessen the amount of cutting required.

Cutting treatments. In the frequent cutting treatment (I) the grass was cut on attaining a height of 8-12 inches, i.e., about six times per year (Table II). The infrequent cutting treatment (II) involved mowing about three times per year, when the grass reached a height of 15-20 inches. Owing to the severity of the competition with the trees (as described later), the plots of treatment II were disc-cultivated in March, 1943, leaving swards only under treatment I. In the years 1941-44, a small motor-operated reciprocating grass cutter was used but this was replaced in 1945 by a tractor-drawn gangmower consisting of three spaced cylindrical mowers. This is shown on one of the grass plots in Fig. 1. Both mowers were arranged to cut the sward to about 2 in., but the gangmower is regarded as being more satisfactory because it cuts the vegetation into small pieces which are spread evenly over the ground and do not lie in dense lumps.

Manurial treatments. A summary of the five manurial treatments, V, W, X, Y, Z, is given in Table II, where N = 1 cwt. (112 lb.) per acre of sulphate of ammonia and P = 1 cwt. per acre of superphosphate. The superphosphate is applied in January and the sulphate of ammonia applications are divided into two equal parts, one applied in February, the other in May.

Treatments W and X were started on mixtures A and B in 1941, one year after sowing, the sub-plots receiving 2½ cwt. and 5 cwt. per acre respectively of sulphate of ammonia. In the following year the two superphosphate treatments Y and Z were

TABLE II.

Cutting and manurial treatments on swards.

	Year.	No. of cuttings.		Manurial treatments on swards.							
		Treatment I " frequent ".	Treatment II " infrequent ".	Mixtures A and B.					Mixture C.		
				V.	W.	X.	Y	Z.	W.	V→X.*	Z.
Period I	1940	0	0	0	0	0	0	0	0	0	0
	1941	7	4	0	2½N	5N	0	0	0	0	0
	1942	5	3	0	2½N	5N	4P	2½N,4P	2½N	0	2½N,4P
Period II	1943	6	Disced	10N	7½N	5N	4P	7½N,4P	7½N	5N	7½N,4P
	1944	4	—	10N	7½N	5N	4P	7½N,4P	7½N	5N	7½N,4P
	1945	7	—	10N	7½N	5N	4P	7½N,4P	7½N	5N	7½N,4P
	1946	5	—	10N	7½N	5N	4P	7½N,4P	7½N	5N	7½N,4P

* V 1940-42, changed to X 1943-46.

started, Z receiving 2½ cwt. per acre of sulphate of ammonia in addition to 4 cwt per acre of superphosphate. At the same time, treatments W and Z were extended to include sub-plots sown with mixture C.

The nitrogen dressings were purposely kept fairly low at first, with the objects of improving fruit colour and of seeing to what extent the clover in the mixtures would supply the required nitrogen. By 1943, when it was seen that many of the trees, especially those under cutting treatment II, were showing signs of nitrogen deficiency, swards of this treatment were turned in, as already mentioned, and plots under treatment I which had previously received the lowest amounts of nitrogen were given greatly increased amounts. Treatments W and Z, previously 2½ cwt. per acre of sulphate of ammonia, were increased to 7½ cwt. per acre, without and with superphosphate as before. An even higher dressing of sulphate of ammonia (10 cwt. per acre) was started on sub-plots V, which had previously received no manure. The 5 cwt. per acre of sulphate of ammonia (X) remained unchanged on mixtures A and B, and was extended to sub-plots of mixture C which had previously received no manure. These manurial treatments established in 1943 were repeated in 1944, 1945 and 1946.

The grass under treatment II received 2 cwt. per acre of sulphate of ammonia in 1943 just before being turned in. No manure was applied on these plots in 1944, but 3 cwt. per acre of sulphate of ammonia was given in 1945 and again in 1946.

The cultivated strip (treatment III) received no manure until 1945, then 3 cwt. per acre of sulphate of ammonia, repeated in 1946.

No potash has been applied at any time in this investigation.

CHANGES IN SWARD COMPOSITION

Changes in composition of the swards were recorded in 1942, 1943, 1944 and 1945 by the percentage area method of pasture analysis (Fenton, 1933). Ten samples were taken in each sub-plot of 192 square yards, using a 20 cm. square grid sub-divided into 100 squares, each of 4 sq. cm. The record was taken in March each year, except in 1943 when the composition was determined in January before discing the grass having the " infrequent " cutting (treatment II).

The sward was analysed into four categories—grasses, clovers, weeds and bare spaces. In general the swards became established satisfactorily in 1940 and were thick and continuous by next March, only small gaps (occupying 6-8 per cent. of the area in 1945) appearing later where the bulky cut material had smothered the underlying vegetation. The weed category, which does not include any grasses (which invaded mixture B in particular), accounted for only about 1 per cent. of the area in 1945. Thus almost the whole area was covered by grasses and clovers. Table III, giving the percentage covered by clovers, indicates the relative abundance of these two categories in the swards resulting from mixtures A and B.

At first mixture B, containing the red clover, had, as would be expected, a very much greater amount of clover than mixture A. By spring, 1942, the red clover which is largely a biennial was beginning to die out and the wild white clover was becoming established. The clovers recorded in mixture B, in 1942, were about half red and half wild white. From 1943 onwards the clovers on all plots were almost entirely wild white. It can be seen in Table III that the plots of mixture B had much more clover in 1942 than those of mixture A, but from 1943 onwards the differences were, in general, small. It was evident by 1943, when the swards of cutting treatment II

TABLE III.

Percentage of clovers in swards produced by different seeds mixtures and by the cutting and manurial treatments.

Treatment.	Sub-plot.	Mixture A.						Mixture B.					
		Period I.			Period II.			Period I.			Period II.		
		Manures.	March 1942.	Jan. 1943.	*New Manures.*	March 1944.	March 1945.	*Manures.*	March 1942.	Jan. 1943.	*New Manures.*	March 1944.	March 1945.
I. Swards frequently cut.	V	*0*	6	36	*10N*	1	0	*0*	28	37	*10N*	4	6
	W	*2½N*	4	7	*7½N*	1	0	*2½N*	4	7	*7½N*	4	9
	X	*5N*	0	0	*5N*	0	0	*5N*	2	1	*5N*	1	5
	Y	*4P*	14	40	*4P*	38	21	*4P*	24	39	*4P*	54	48
	Z	*2½N 4P*	12	11	*7½N 4P*	0	0	*2½N 4P*	20	20	*7½N 4P*	6	9
II. Swards infrequently cut.	V	*0*	16	21	Sward disced in March, 1943			*0*	30	20	Sward disced in March, 1943		
	W	*2½N*	3	6				*2½N*	7	7			
	X	*5N*	0	0				*5N*	4	3			
	Y	*4P*	12	27				*4P*	33	26			
	Z	*2½N 4P*	9	11				*2½N 4P*	30	20			

were disced in, that this infrequent cutting resulted in a sward containing less clover than that resulting from frequent cutting. These differences, however, are slight compared with the differences between manurial treatments and it is apparent from the data of Table III that, in determining the ultimate clover content of the sward, the manurial treatment in this case has been a much more important factor than either the proportion of clover in the seeds mixture or the frequency of cutting.

Considering the effects of the various manurial treatments in detail, addition of sulphate of ammonia was always followed by a decrease in the clover content. Sub-plots V contained a high proportion of clover in 1943, having received no nitrogen until that year, but addition of 10 cwt. per acre of sulphate of ammonia then resulted in almost complete extinction of the clover in one year. In the case of sub-plots W, 2½ cwt. per acre of sulphate of ammonia in 1941 was followed by a record of only 4 per cent. clover in March, 1942, increasing to 7 per cent. in 1943 after a further 2½ cwt. per acre ; increase of the rate of application to 7½ cwt. per acre then resulted in disappearance of clover in 2 years in swards A, and prevented any increase in the proportion of clovers in swards B. Application of 5 cwt. per acre of sulphate of ammonia (sub-plots X) prevented the appearance of any clover in sward A, and caused the proportion in sward B to be even lower than in the case of manurial treatment W.

The data for sub-plots Y (superphosphate only) show that a high proportion of clover can be maintained in the absence of added nitrogen. The 1943 figures are slightly higher than those for sub-plots V, which until then had received no manures, but evidently the addition of superphosphate to this already phosphate-rich soil did not markedly encourage the clover at the expense of the grass. Unfortunately from 1943 onwards there was no control treatment receiving no manures. The addition of superphosphate on sub-plots Z did not prevent a decrease in the proportion of clovers associated with applications of sulphate of ammonia as for treatment W, but no rigid comparison between W and Z can be made because sub-plots W received nitrogen a year before sub-plots Z.

Thus the application of 5 cwt., 7½ cwt. or 10 cwt. per acre of sulphate of ammonia caused the complete disappearance of clover from swards A, and a reduction of the percentage of clover to 5-9 per cent. in swards B by 1945, even when superphosphate was added as well. Similar results were obtained for mixture C, where all sub-plots received 5 cwt. or 7½ cwt. per acre of sulphate of ammonia, yielding swards containing 6-13 per cent. of clover in 1945. On the other hand, in the absence of added nitrogen but with superphosphate, swards rich in clover were obtained and maintained. Decrease in the proportion of clovers was always associated with vigorous growth of grasses, for instance the cocksfoot in mixture A grew more vigorously than the grasses of mixture B. (The 1945 swards on sub-plots V, W, X and Z of mixture A were composed almost entirely of cocksfoot.) Further, the infrequently cut swards grew more vigorously than those cut frequently, and of all the manurial treatments Y (superphosphate only) produced least growth. These facts are all in agreement with the theory that disappearance of clover is caused by increased competition of the grasses under certain conditions, but a possible direct harmful effect of sulphate of ammonia on the clovers is not ruled out by these data. The problem has been the subject of further study, the results of which are published in Paper V of this series (p. 271).

Because of the known beneficial effects of leguminous plants, it appears desirable to maintain a high proportion of clover in the sward. These results indicate that this may not be possible without withholding sulphate of ammonia. In the absence of data relating to the amount of nitrogen fixed by the clover, the choice between added nitrogen in the absence of clover on the one hand and clover-produced nitrogen on the other, depends on the reaction of the tree (see later). The possibilities of using other inorganic sources of nitrogen applied at different seasons are considered elsewhere. (Rogers, Raptopoulos and Greenham, Paper V of this series. See p. 271.)

EFFECT OF SPRAYING ON THE SWARD.

The trees received the usual spray programme, including tar oil plus petroleum oil or dinitrocresol ; the swards thus received spray drifts comparable with those in normal commercial orchards except for a few small strips which were covered during spraying in some years for observation purposes. Unlike the severe damage caused by the winter spraying on annual cover crops (Rogers and Raptopoulos, 1946b), no appreciable scorching was observed on these swards except for a temporary yellowing of the foliage when compared with the protected strips. A mild check to growth resulted but the plants were soon indistinguishable in height from those which had been protected.

COVER CROP YIELDS.

Methods of determination.

The swards were not mown during the year of sowing (1940), and became well established. The first cut was made in May, 1941, and in subsequent years mowing generally commenced about 20th April, though the exact date depended on the amount of growth made by the swards. The date of the last cut varied between 27th July and 18th October, depending again on the amount of growth. In the last two years (1945 and 1946) it was decided not to mow after harvesting the apples, because it was considered that at that time of year competition cannot harm the tree and might have beneficial effects in hastening maturing of the shoots. Within these limits, cuttings were made at the times when the grass reached the prescribed heights, as described above. The grass was cut before seeding even in the infrequent cutting treatment, but the formation of some seed was unavoidable in inaccessible places close to the trees.

Immediately before each mowing, a sample strip 36 ft. by 3 ft. was cut from each sub-plot by the reciprocating grass cutter. The cut material from each strip was weighed and redistributed over the strip after taking a sub-sample of about 100 gm. in an airtight tin. Each sub-sample was weighed and dried to a constant weight at 100° C. in an electric oven, for determination of dry matter content.

Total yields.

The weights of cut material, calculated in tons per acre both fresh weight and dry weight, are given in the tables. Table IV shows the data for 1941 and 1942. Table V shows the data for 1943-46, i.e., for the period subsequent to the changes in manurial treatments, only the " frequent " cutting treatment (I) being represented in this period.

Table IV.

Weights (tons/acre) of cut material in period I (1941-42) produced by different seeds mixtures, cutting treatments and manurial treatments.

Mixture.	Sub-plot.	Manures.	Treatment I. Swards frequently cut.				Treatment II. Swards infrequently cut.			
			1941.		1942.		1941.		1942.	
			Fresh.	*Dry.*	Fresh.	*Dry.*	Fresh.	*Dry.*	Fresh.	*Dry.*
A	V	0	1·73	*0·44*	2·60	*0·63*	3·76	*0·92*	6·72	*1·48*
	W	2½N	3·77	*0·94*	3·66	*1·03*	6·46	*1·65*	6·44	*1·52*
	X	5N	6·19	*1·27*	6·47	*1·59*	9·59	*2·14*	10·01	*2·09*
	Y	4P			3·93	*0·90*			6·27	*1·27*
	Z	2½N 4P			5·06	*1·40*			7·93	*1·73*
	Mean		**3·90**	**0·88**	**4·34**	**1·11**	**6·60**	**1·57**	**7·47**	**1·62**
B	V	0	2·88	*0·61*	4·89	*1·03*	7·95	*1·88*	8·97	*1·51*
	W	2½N	3·96	*0·85*	3·21	*0·84*	8·74	*1·96*	8·72	*1·69*
	X	5N	5·88	*1·19*	5·16	*1·27*	9·79	*2·07*	11·50	*2·23*
	Y	4P			4·33	*0·87*			9·64	*1·61*
	Z	2½N 4P			4·56	*1·03*			10·91	*1·81*
	Mean		**4·24**	**0·88**	**4·43**	**1·01**	**8·83**	**1·97**	**9·95**	**1·77**
C	V	0			1·32	*0·30*				
	W	2½N			2·70	*0·63*				
	Z	2½N 4P			1·89	*0·48*				
	Mean				**1·97**	**0·47**				

Period I (1941-42) (Table IV).

Considering all manurial treatments on both mixtures A and B together for this period, treatment II (" infrequent " cutting) gave a mean yield of 8·22 tons fresh weight per acre per year compared with 4·20 tons for treatment I (" frequent " cutting). The corresponding dry weight figures are 1·76 tons per acre per year for treatment II, and 0·97 tons for treatment I. The difference between yields for cutting treatments is strongly significant in both fresh and dry weight. This effect is undoubtedly due to the larger surface available for photosynthesis, considering the season as a whole, on an infrequently cut sward and shows the popular view, that frequent cutting increases the growth, to be a fallacy.

Considering both cutting treatments and all manurial treatments together for the same period, mixture A gave a mean fresh weight yield of 5·62 tons per acre per year, significantly less than that for mixture B (6·80 tons). The corresponding dry weight yields were not significantly different. Considering the fresh weight yield data again, the cutting × mixture and mixture × manure interactions were significant, the difference between mixtures A and B being diminished or eliminated by short cutting or by additions of sulphate of ammonia. To illustrate the latter point, the mean fresh weights (tons per acre per year) for mixtures and manurial treatments separately, considering both cutting treatments together, are as follows :

Manures.	*Mixture A.*	*Mixture B.*	*Difference.*
V (0)	3·70	6·17	2·47
W ($2\frac{1}{2}$N)	5·08	6·16	1·08
X (5N)	8·06	8·08	0·02
		Significant difference	1·24

Thus mixture B gave a fresh weight yield significantly greater than that for mixture A when no manure was added, but this difference was reduced to being not quite significant at P = 0·05 when sulphate of ammonia was added at $2\frac{1}{2}$ cwt. per acre and almost disappeared altogether for twice this application.

Considering these data in relation to those for percentage of clover (Table III), it seems clear that the lessening of the differences between the sward yields, with increasing amount of nitrogenous manure, is largely connected with the elimination of clover under these conditions.

The 1941-42 data show differences between yields corresponding to manurial treatments, considering both cutting treatments and both mixtures A and B together as follows :

	Tons per acre per year.	
Manures.	*Fresh weight.*	*Dry Weight.*
V (0)	4·94}	1·06
W ($2\frac{1}{2}$N)	5·62}	1·32
X (5N)	8·07	1·73
Significant difference	0·88	0·21

All differences are significant except that the fresh weight corresponding to an application of $2\frac{1}{2}$ cwt. per acre of sulphate of ammonia is not quite significantly greater at P = 0·05 than that when no manure was added. As would be expected, growth was markedly stimulated by the application of nitrogen. Manurial treatments Y and Z are not considered in this comparison, because the first applications were not made until 1942.

Period II (1943-46) (Table V).

During this period, the manurial treatments were different from those in 1941-42 and only the " frequent " cutting treatment (I) was represented. Considering all manurial treatments together, the fresh and dry weights for mixtures A and B are as follows :

	Tons per acre per year.		
	Mixture A.	*Mixture B.*	*Significant difference.*
Fresh weight	6·49	5·94	0·39
Dry weight	1·39	1·22	0·09

Thus, while mixture A gave a significantly lower fresh weight yield than mixture B in 1941-42, its yield, both fresh and dry, was significantly higher than that for mixture B in 1943-46. This change is due to changes in sward composition ; for instance, the yields of mixture B in 1941 were considerably influenced by the vigorous growing red clovers, which later disappeared from the sward. The greater yield of mixture A in 1943-46 is doubtless due to the increasing dominance of the vigorous cocksfoot.

TABLE V.

Weights (tons/acre) of cut material in period II (1943-46) produced by swards under treatment I (frequent cutting).

Mixture.	Sub-plot.	Manures.	1943.		1944.		1945.		1946.		TOTAL.	
			Fresh.	*Dry.*	Fresh.	*Dry.*	Fresh.	*Dry.*	Fresh.	*Dry.*	Fresh.	*Dry.*
A	V	10N	10·56	*2·30*	6·25	*1·23*	6·55	*1·81*	7·22	*1·24*	30·58	*6·58*
	W	7½N	8·69	*2·01*	5·83	*1·17*	6·43	*1·64*	7·66	*1·34*	28·61	*6·16*
	X	5N	7·57	*1·77*	4·73	*0·98*	4·85	*1·26*	6·88	*1·21*	24·03	*5·22*
	Y	4P	6·06	*1·25*	2·54	*0·55*	3·14	*0·79*	5·56	*0·96*	17·30	*3·55*
	Z	7½N 4P	9·55	*2·12*	6·20	*1·24*	5·98	*1·55*	7·57	*1·30*	29·30	*6·21*
	Mean		**8·49**	**1·89**	**5·11**	**1·03**	**5·39**	**1·41**	**6·98**	**1·21**	**25·96**	**5·54**
B	V	10N	9·23	*1·93*	6·73	*1·27*	5·81	*1·41*	7·62	*1·29*	29·39	*5·90*
	W	7½N	7·82	*1·84*	5·70	*1·08*	4·94	*1·17*	7·23	*1·22*	25·69	*5·31*
	X	5N	5·44	*1·32*	4·19	*0·93*	3·64	*0·87*	6·64	*1·12*	19·91	*4·24*
	Y	4P	5·59	*1·22*	3·19	*0·72*	3·00	*0·70*	5·77	*0·96*	17·55	*3·60*
	Z	7½N 4P	7·34	*1·69*	5·62	*1·13*	5·21	*1·22*	8·05	*1·36*	26·22	*5·40*
	Mean		**7·08**	**1·60**	**5·09**	**1·03**	**4·52**	**1·07**	**7·06**	**1·19**	**23·75**	**4·89**
C	W	7½N	8·14	*1·78*	4·01	*0·84*	5·04	*1·27*	6·19	*1·02*	23·38	*4·91*
	X	5N	7·65	*1·64*	3·14	*0·66*	4·33	*1·04*	8·27	*1·44*	23·39	*4·78*
	Z	7½N 4P	7·41	*1·74*	4·14	*0·91*	5·25	*1·37*	7·65	*1·23*	24·45	*5·25*
	Mean		**7·73**	**1·72**	**3·76**	**0·80**	**4·87**	**1·23**	**7·37**	**1·23**	**23·74**	**4·98**

The data for mixture C show that growth was little in 1942, but comparison of the means for 1943-46 with those for corresponding treatments (W, X and Z) on mixtures A and B shows that the yield from C was about the same as that from B :

Tons per acre per year.

	Mixture A.	*Mixture B.*	*Mixture C.*
Fresh weight	6·83	5·99	5·94
Dry weight	1·47	1·25	1·25

No statistical analysis is possible with regard to mixture C, because the plots were not in the main experiment.

Considering grass mixtures A and B together, the mean yields for the various manurial treatments are as follows :

Tons per acre per year.

Manures.	*Fresh weight.*	*Dry weight.*
Y (4P)	4·36	0·89
X (5N)	5·49	1·18
W (7½N)	6·79	1·43
Z (7½N, 4P)	6·94	1·45
V (10N)	7·50	1·56
Significant difference (P = 0·05)	0·62	0·15

All differences except those included within brackets are significant. Thus the effects of increasing applications of sulphate of ammonia from 0 to 5 cwt. and from 5 to 7½ cwt. per acre are both significant. Yields were further increased, though not significantly as regards dry weight, by increasing this application from 7½ cwt. to 10 cwt. per acre. Comparison of sub-plots W and Z shows that addition of superphosphate had no significant effect on the yield of cover crop in presence of 7½N ; but it appeared to be effective in 1942 with 2½N, when much clover still remained in the sward (Tables III and IV).

Comparison of sub-plots Y and X shows that the application of 5 cwt. per acre of sulphate of ammonia instead of 4 cwt. per acre of superphosphate increased the fresh weight yield by 26 per cent. and the dry weight yield by 33 per cent. Use of a further 2½ cwt. per acre of sulphate of ammonia increased the yields by a further 24 per cent. (fresh) and 21 per cent. (dry), while yet another 2½ cwt. per acre increment corresponded to further yield increases of only 10 per cent. (fresh) and 9 per cent. (dry). If the cover crop yield were the only criterion, therefore, there would seem to be little point in applying sulphate of ammonia at rates exceeding 7½ cwt. per acre ; the effects of these manurial treatments on the trees are, however, considered later.

SEASONAL DISTRIBUTION.

Fig. 3 shows, in diagrammatic form, the amount of fresh material cut at each mowing during three typical years—1941, 1943 and 1945. The amounts are in tons per acre, representing the mean of all plots sown with mixtures A and B, with the "frequent" cutting treatment.

It will be seen that in 1941 (the first year after sowing) the maximum yield was in August but subsequently, in 1943 and 1945, the bulkiest cuts were made in May (and the same is true of 1942 and 1944). In 1946, when April was unusually dry and June very wet, the heaviest yield was in June ; but if all six years are considered together, over half the total material was cut by the end of May. Growth of the sward decreased in the summer, but there is some evidence of an increase in growth (an "aftermath") in September. As mentioned earlier, in the last two seasons (1945 and 1946) the policy was adopted of allowing the grass to grow uncut after the end of July, as a certain amount of competition for water at that time of year might assist in the ripening of both fruit and young wood.

COMPARISON WITH OTHER TYPES OF COVER CROP.

The experimental trial of annual cover crops (Rogers and Raptopoulos, 1946a, Table II) showed yields ranging from 0·63 to 1·19 tons dry matter per acre per year, these figures being the mean of the values for the seasons 1941, 1942 and 1943. The trial of short-term leys (Rogers and Raptopoulos, 1945, Table I) showed yields ranging from 0·22 (weeds) to 1·48 (grass and clover) tons dry matter per acre per year over the seasons 1941 and 1942. The mean yields for all seeds mixtures and all manurial treatments in the present experiment for the years 1943-46 are shown in Table V to range from 0·89 to 1·65 tons dry matter per acre per year. In terms of amount of material supplied to the soil per year by the cover crop, therefore, the long-term leys, even when kept cut short, appear to be at least as efficient as annual cover crops and short-term leys.

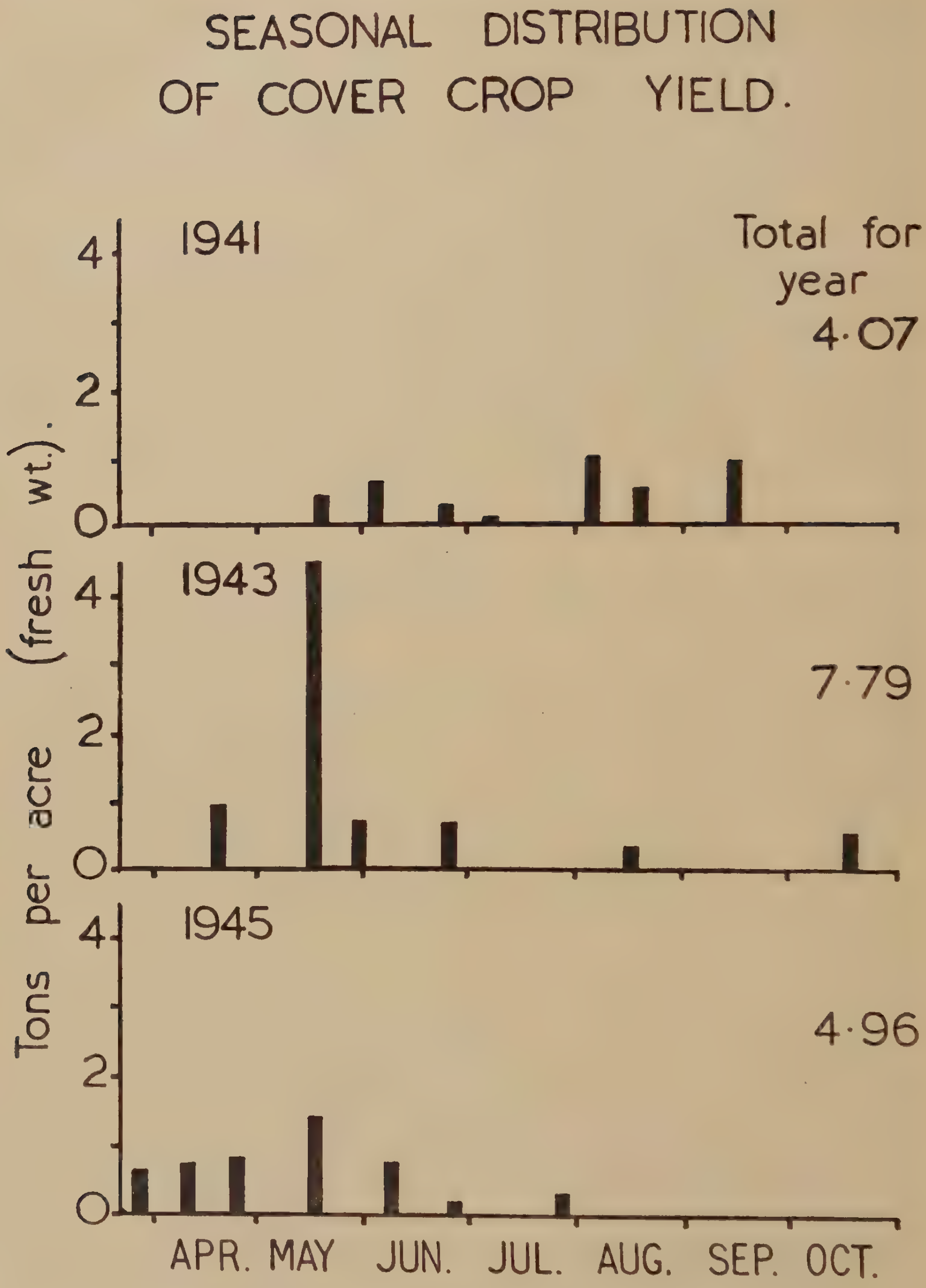


FIG. 3.

Changes in seasonal distribution of cover crop yields.

EFFECT OF COVER CROP ON AIR TEMPERATURES.

It is an established meteorological fact that on "radiation nights" the temperature immediately above grass tends to be lower than that immediately above bare soil (Cornford, 1938).

Preliminary comparisons of the minimum temperatures at a height of 5 ft. 6 in. during the blossom season were made, with thermometers in duplicate and triplicate, on the cultivated strip and adjoining grass strips in 1941 and 1943. These showed that at this height the difference in minima is only of the order of 1° F. over long grass, and less over short grass.

In subsequent years, temperature studies were continued on another plantation, in which larger areas of grass and cultivated ground were available. These fuller data, which will be reported separately, confirm that the difference in minima is generally less than 1° F. Thus the risk of frost damage is not materially increased by grassing down but nevertheless it should be borne in mind and minimized by keeping the grass cut as short as possible during the blossom season.

EFFECT OF COVER CROP ON THE SOIL.

Organic matter and nitrogen content.

Organic matter content was determined at Wye College by Dr. N. H. Pizer, using the method of Walkley, on some samples taken on 28th June, 1940. Values obtained were about 3 per cent. for the 0-6 in. range of depth and about 1·5 - 2 per cent. for the whole range examined (0-24 in.).

A series of soil samples was taken on 12th October, 1943, from all of the 30 sub-plots on which swards of cutting treatment II had been turned in in March of that year, and from the 10 continuously cultivated sub-plots. Organic matter content (Walkley) and nitrogen content (Kjeldahl modified by Ashton) were determined at East Malling. They showed considerable variation, none of the differences being significant.

The 1940 samples were not taken from each sub-plot; the sampling unit in that year was the whole plot, i.e., the strip of five sub-plots each receiving the same seeds mixture and cutting treatment but differing in manurial treatment. Moreover the series was incomplete in that the whole area of the trial was not sampled. It is not therefore possible to make a comprehensive study from these data of the changes in soil organic matter content during the period 1940-43. There are indications, however, of a slight decrease, both in the soil which had been cultivated throughout and in the soil on which a ley had been grown and turned in. This type of result was obtained by McHenry *et al* (1947) in a 9-year investigation in Nebraska, where soil which had been under 3-year leys showed losses in organic matter roughly equal to the loss under continuous cultivation. Such losses may be due to a seasonal fluctuation quite secondary in importance to a general long-term upward trend under grass swards.

The building-up of the organic matter content of the soil by the use of grass swards may be a very slow process, and the beneficial effect of a ley cannot be expressed solely in terms of the change brought about in the soil organic matter content, for there may well be improvements in soil structure resulting from the activity of micro-organisms sustained and stimulated by the provision of decomposible plant residues as "energy material" (McCalla, 1947). It is recognized that the most stable soil aggregates are formed under a grass sward where the extensive fibrous roots are continually dying and being replaced, providing a continual source of " energy material ", in addition to the cut grass lying on the surface, for the microbial population. The rapid disappearance of organic matter supplied by a ley, as occurred in this investigation, may be an indication of beneficial microbial activity in the soil, ensuring a supply of readily available nutrients and improving the soil structure. Further work is needed on the effect of leys on microbiological activity and on soil aggregation.

Waksman (1936) has pointed out that the C/N ratio in a soil provides an indication of the stage of decomposition of its organic matter, high ratios such as 20 : 1 showing that little decomposition has occurred and low ratios (10 : 1 or less) showing an advanced stage of decomposition. Determinations made on the 1943 soil samples showed C/N ratios ranging from 7·9 to 10·3, confirming the view that the organic matter supplied by the ley was in an advanced stage of decomposition seven months after it was incorporated in the soil. Microbiological activity must have been considerable and the failure to demonstrate any increase in organic matter content is therefore not surprising.

Soil density.

Improvement in soil structure, whether due directly to incorporation of organic matter or indirectly to the action of micro-organisms in increasing aggregation, should result in a less compact soil with improved aeration and water retention capacity, providing better conditions for plant growth. An attempt was made to compare the density or compactness of the soil after turning in the swards in 1943, with that of the soil which had been cultivated throughout.

The instrument used for this purpose was similar to the soil compactometer described by Heath (1934). Its essential feature is a cylindrical probe, 13 mm. in diameter, with a pointed end, which is driven into the soil by the impact of a 3 kg. weight falling from a constant height of 16 cm., the depth of penetration being recorded at each impact.

Determinations were made in early April, 1945, on all sub-plots E, F and G, i.e., 10 sub-plots each of treatments cultivated throughout (III), grass throughout (I) and grass turned in (II). The sub-plots of treatment II had been cultivated in exactly the same way as those of treatment III during the two years since turning in and, at the time of determination, the soil surface was level and weatherbeaten as spring cultivations had not started. The top 2 cm. was discarded in all cases so that the compactometer might stand firmly, and all depths were reckoned from this adjusted soil level. Readings were taken to a depth of about 30 cm.

Fig. 4 shows graphically the depth reached at each impact. Each point through which the curves are drawn represents the mean of all ten determinations for that particular treatment. For any given number of impacts, the probe penetrated least on grass and most on turned-in grass, the cultivated treatment being intermediate. Determinations of soil moisture content on samples taken at the same time showed closely similar values for all three treatments, so that differences in penetration cannot be attributed to any differences in soil moisture content.

The slope of each curve (Fig. 4) gives an estimate of the resistance at any given depth in terms of number of impacts per centimetre. The appropriate values have been deduced from Fig. 4 and are presented graphically as Fig. 5. Here it is seen that at all depths to 24 cm. treatment II showed least resistance and I most, III being intermediate. The differences in resistance are, however, slight below 12 cm., the contrast between treatments in the 0-12 cm. zone being much greater.

As it was not possible to make determinations on the whole area of the trial, the results can be interpreted statistically only if the assumption be made that the soil was uniform in respect of compactness between strips E, F and G at the start of the trial. This assumption would seem to be reasonable. Statistical analysis shows significant differences between all three strips in the number of impacts needed to drive the probe 12 cm. into the soil, values being :

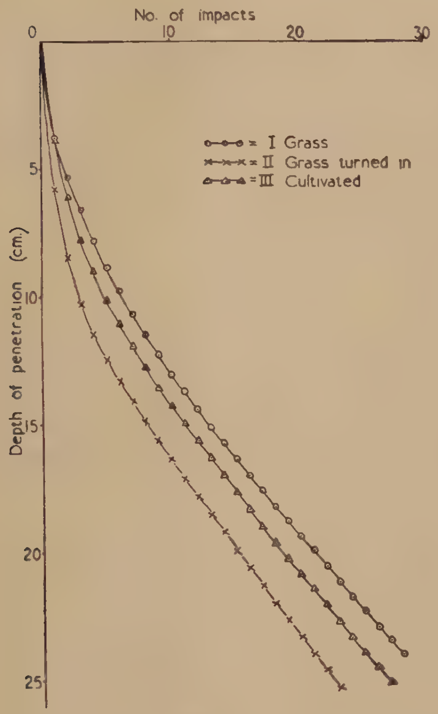


Fig. 4.

Soil hardness measured by the compactometer. Depth of penetration by the probe at each successive impact.

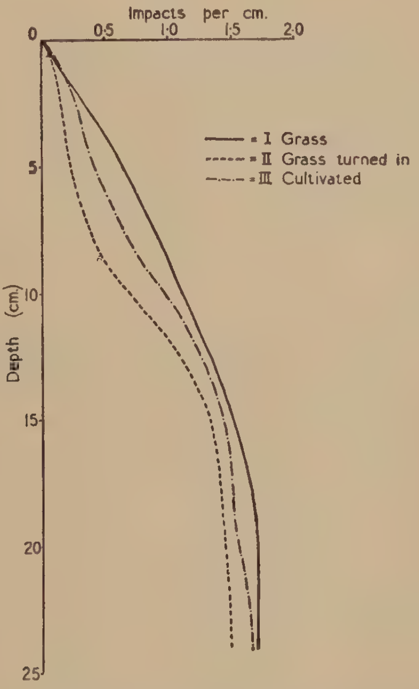


Fig. 5.

Soil hardness measured by the compactometer. Resistance (in impacts per centimetre) to penetration by the probe, at different depths.

Treatment II—Grass turned under	4·8
Treatment III—Cultivation	7·7
Treatment I—Grass	9·4

If the number of impacts needed to drive the probe 30 cm. into the soil be considered, the value for treatment II is still significantly less than those for the other treatments, but there is then no significant difference between cultivation and grass :

Treatment II—Grass turned under	30·7
Treatment III—Cultivation	36·5
Treatment I—Grass	39·6

As seen in Fig. 5, the difference is large only to a depth of 12 cm., and the effect of adding a further 18 cm. is simply to tend to mask the significance of the differences.

The incorporation of a 3-year ley therefore decreased the compactness of the soil, the effect being considerable as long as two years after incorporation. There was a smaller difference between continued grass and cultivation, showing the soil under grass to be rather more resistant to penetration of the probe than that receiving regular cultivations throughout. The extra resistance, in this case, is more likely to be due to the living grass roots than to any difference in soil structure.

SOIL MOISTURE.

The variations in soil moisture on certain plots were recorded by the use of soil moisture tensiometers (Rogers, 1935). The complete results will be published separately, but the main conclusion was that the soil under grass began losing water 15-30 days earlier in the spring than did cultivated soil, as noted in another trial (Rogers, 1937). On the other hand, at least in some cases, the soil under grass was more easily replenished by the heavy rains of summer and early autumn.

EFFECT OF COVER CROP ON THE TREES.

GROWTH.

Girth. The principal method used to estimate the effect of the swards on the growth of the trees was the standard annual measurement of girth of the trunk at a marked height on each tree, the measurements being subsequently converted into cross-sectional area, assuming the cross-section to be circular. Table VI gives a summary of the mean percentage increments in cross-sectional area for the three growing seasons 1940-42, for the four growing seasons 1943-46, and for all seven seasons together.

In statistical analysis of these data, the variate analysed was, for each sub-plot of three trees, the sum of the increments in the logarithms of the areas of cross-section over the period concerned. For the seasons 1940-42, before any of the swards were turned in, the small differences between cutting treatments were not significant, neither was the difference significant between either cutting treatment and clean cultivation, but mixture B gave significantly greater increments than mixture A. The lack of significance in the difference between grass (values for all treatments ranging from 5·1 per cent. to 9·7 per cent.) and cultivation (12·2 per cent.) is somewhat surprising and is a reflection on the large errors involved in the use of such variable trees. In an attempt to eliminate error due to initial variation between trees (including that due to the absence of a uniform rootstock throughout), the statistical analysis was repeated, covariancing on to a corresponding variate representing girth increments from 1934 to 1939, but none of the differences was found to be significant.

Considering further the 1940-42 data, however, it is of interest to examine the three seasons separately. It is seen then that while the trees on the cultivated plots increased their cross-sectional area by about 4 per cent. each year, those on the grass plots began to show marked signs of a check in 1941. A repetition of the statistical analysis, omitting season 1940, showed that for the seasons 1941 and 1942 considered together, both mixtures A and B gave significantly lower increments than cultivation, the check being significantly greater with mixture A than with mixture B but there

Percentage increase in cross-sectional area of trunk.

Treatment.	Mixture.	Sub-plot.	Period I.					Period II.						1940-6 inclusive.
			Manures.	1940.	1941.	1942.	1940-2 inclusive.	*New Manures.*	1943.	1944.	1945.	1946.	1943-6 inclusive.	
I. Swards frequently cut.	A	V	*o*	3·5	1·4	1·0	6·0	*10N*	0·9	4·6	4·5	3·0	13·7	20·5
		W	*2½N*	2·6	1·4	2·0	6·1	*7½N*	1·0	4·7	3·1	2·9	12·0	19·0
		X	*5N*	4·0	1·2	2·0	7·4	*5N*	1·0	3·2	4·5	2·3	11·5	19·8
		Y	*4P*	2·7	1·9	1·5	6·3	*4P*	1·1	2·3	2·4	2·3	8·3	15·0
		Z	*2½N* *4P*	2·2	1·3	1·5	5·1	*7½N* *4P*	0·6	3·3	4·0	2·9	11·9	17·6
		Mean		**3·0**	**1·4**	**1·6**	**6·2**		**0·9**	**3·6**	**3·7**	**2·7**	**11·6**	**18·4**
	B	V	*o*	4·2	2·8	2·5	9·7	*10N*	2·0	7·2	5·5	3·4	19·5	31·1
		W	*2½N*	2·8	2·3	2·0	7·3	*7½N*	1·6	5·1	4·8	3·5	15·8	24·2
		X	*5N*	3·8	2·1	2·2	8·3	*5N*	2·3	5·1	3·7	3·3	14·9	24·5
		Y	*4P*	3·1	1·8	1·3	6·4	*4P*	1·6	2·5	3·4	3·3	11·8	18·9
		Z	*2½N* *4P*	3·1	1·7	1·4	6·4	*7½N* *4P*	1·1	5·1	6·8	3·3	17·5	25·0
		Mean		**3·4**	**2·1**	**1·9**	**7·6**		**1·7**	**5·0**	**4·8**	**3·4**	**15·9**	**24·7**
	C	W	*2½N*	3·7	3·8	1·5	9·2	*7½N*	1·1	5·1	4·8	2·2	14·0	24·5
		V → X	*o*	3·4	2·6	2·0	8·3	*5N*	0·7	5·1	2·8	3·6	13·1	22·5
		Z	*2½N* *4P*	2·2	2·5	0·8	5·5	*7½N* *4P*	0·5	3·1	2·8	2·7	9·1	15·3
		Mean		**3·1**	**3·0**	**1·4**	**7·7**		**0·8**	**4·4**	**3·5**	**2·8**	**12·1**	**20·8**
II. Swards infrequently cut.	A	V	*o*	3·7	1·6	0·9	6·4	All cultivated, with uniform N.	1·0	8·2	6·0	4·1	20·6	28·2
		W	*2½N*	3·1	1·6	0·9	5·6		0·6	6·9	5·0	3·1	17·4	23·9
		X	*5N*	3·6	1·4	0·7	5·8		1·5	7·4	5·5	2·8	18·5	25·3
		Y	*4P*	4·6	1·9	0·6	7·3		1·2	8·4	5·5	3·6	19·9	28·5
		Z	*2½N* *4P*	3·7	1·9	0·8	6·6		0·6	8·2	5·5	3·6	19·0	26·8
		Mean		**3·7**	**1·7**	**0·8**	**6·3**		**1·0**	**7·8**	**5·5**	**3·4**	**19·1**	**26·5**
	B	V	*o*	4·2	2·2	2·0	8·6		1·9	9·2	5·4	3·7	21·7	32·3
		W	*2½N*	4·2	2·2	1·8	8·2		1·5	8·9	5·8	3·9	22·2	32·3
		X	*5N*	5·0	2·2	2·1	9·7		2·8	11·3	5·0	5·3	26·6	38·9
		Y	*4P*	3·7	1·4	1·7	6·9		2·2	8·5	3·9	3·8	19·7	27·9
		Z	*2½N* *4P*	3·5	1·9	2·7	8·2		1·1	9·3	5·5	2·7	20·2	30·2
		Mean		**4·1**	**2·0**	**2·1**	**8·3**		**1·9**	**9·4**	**5·1**	**3·9**	**22·1**	**32·3**
III. Cultivated				**3·9**	**3·7**	**4·0**	**12·2**		**3·1**	**6·7**	**5·5**	**2·2**	**18·4**	**32·9**

was still no significant difference between cutting treatments. The figures for 1942 show the most striking differences :

Frequent cutting (I), mixture A	1·6 per cent.
Frequent cutting (I), mixture B	1·9 ,, ,,
Frequent cutting (I), mixture C	1·4 ,, ,,
Infrequent cutting (II), mixture A	0·8 ,, ,,
Infrequent cutting (II), mixture B	2·1 ,, ,,
Cultivated (III)	4·0 ,, ,,

Statistical analysis on the 1942 data above showed again that both mixtures A and B checked growth significantly compared with cultivation and that the check was significantly greater with mixture A than with mixture B.

Clearly the check to growth in the infrequently cut mixture A (which was by 1942 almost pure cocksfoot) was serious and indeed in some cases branches were dying off. It was on this account that the infrequently cut plots were disced in, in 1943. The continued lack of significance in the difference between cutting treatments is probably due to the fact that frequency of cutting appears to have made no difference to the checking of the trees in mixture B, which lacked the vigorous growing cocksfoot and, at this stage, still had much clover. For the sake of uniformity, however, the infrequently cut plots of this mixture were disced as well.

The right hand part of Table VI shows the percentage increase in cross-sectional area for the seasons 1943-46, during which time the various manurial treatments continued on the grass subjected to frequent cutting, while the disced-in plots (formerly infrequent cutting) received applications of nitrogen uniform with those on the area which remained cultivated throughout. Statistical analysis of these data showed that the disced-in plots gave significantly higher increments than the plots continuing in grass but neither is significantly different from cultivated. Only the disced-in plots showed any marked difference in girth in this period compared with seasons 1940-42. Further, considering the remaining grass plots in the period 1943-46, mixture B gave a significantly higher value than mixture A, where the cocksfoot was presumably competing severely with the trees.

Consideration of the data for individual seasons of the 1943-46 period reveals another example of the delay in the trees' response to changes in cultural methods. In the same way that no marked check to growth occurred in 1940, following sowing of the seeds mixtures in April of that year, similarly relatively little acceleration of growth occurred on the disced-in plots in 1943 after discing in February. In 1944, however, the stimulating effect of turned-in sod was striking :

Frequent cutting (I), mixture A	3·6 per cent.
Frequent cutting (I), mixture B	5·0 ,, ,,
Frequent cutting (I), mixture C	4·4 ,, ,,
Infrequent cutting (II), mixture A (disced in 1943)	7·8 ,, ,,
Infrequent cutting (II), mixture B (disced in 1943)	9·4 ,, ,,
Cultivated (III)	6·7 ,, ,,

Statistical analysis of these data shows that the disced-in sward B stimulated growth significantly more than A, which did not differ significantly from cultivated. Meanwhile, on the continued swards mown frequently, mixture A still checked the trees significantly compared with B, which did not differ significantly from cultivated.

No significant differences were found between the various manurial treatments in the seasons 1940-42 but, in 1943-46, treatment Y gave a significantly lower value than the others. The trees showed reddish bark and hardly any new shoots. Evidently the presence of clover in sub-plots Y did not make up for the lack of added nitrogen. Nitrogen as specified was added in all other treatments, which showed no significant differences between each other.

Considering the figures for total growth over the whole period shown in the right hand column of Table VI, it can be seen that none of the trees remaining under grass, even with the increased nitrogen applications from 1943 onwards, had made as much growth as those under cultivation. The trees under sward A turned-in in 1943 also had not made up for the severe initial check caused by the vigorous cocksfoot. However, the trees under sward B, under treatments V, W and X which had not caused such a severe initial check, more than made up for this check, when the sward was turned in. In the case of treatment X, in which 5 cwt. of sulphate of ammonia was given in 1941 and 1942 before the sward was turned in, and nitrogen similar to the cultivated plots thereafter, the trees exceeded the growth of the cultivated ones, having 38·9 per cent. increase compared with 32·9 per cent. It is not possible to say if this difference is significant.

Some of the main contrasts in growth may be seen most clearly in the upper parts of Figs. 6 and 7, which show the annual increments between 1940 and 1946 in cross-sectional area of the trunk. It can be seen (Fig. 6) that the swards imposed no check on growth in the year of their sowing, but a marked check was given in the following year and continued in 1942. On turning-in the infrequently cut sward (Fig. 7), no marked stimulation occurred immediately in 1943 but a very large increase of growth is seen in 1944, the growth of the trees under treatment II then greatly exceeding that of those under cultivation. The increase of tree growth under all treatments in that year corresponds with destruction of most of the fruit crop by frost, a correlation previously noted by Hoblyn and Bane (1934) and by Wilcox (1937). In 1945, when a good crop of fruit was borne by all trees, growth was more similar under all treatments. In 1946 the trees in frequently cut swards of mixture B (Fig. 6), having shown a continued check ever since 1941, surpassed the cultivated ones for the first time. The crossing of the lines at this period is of considerable interest. At this time also, the swards receiving phosphate but no nitrogen (Fig. 6) produced tree growth equal to that of swards with nitrogen, after showing a greater check ever since 1941. Meanwhile sward A (Fig. 7) still showed a check compared with B, as in every other season except 1945.

Pruning weights. With the spur-pruning method used on these trees, the weight of prunings cut every year represents fairly well the new shoot growth of the preceding season. This weight of new wood was recorded every year except 1942, and the summarized results appear in Table VII.

These results show that the various treatments had much the same general effect on new wood production as they had on increase of cross-sectional area of the trunk. In 1941 all grass treatments showed less new wood than did the cultivated plots, and visual observations showed that new growth was again restricted under grass in 1942. The disced-in plots showed new wood comparable to that in cultivation in 1944 (but not in 1943, the year of discing) but, considering together the four seasons 1943-46, no treatments (continued grass or erstwhile sward) showed quite as much growth as did the cultivated plots. Clearly, therefore, the disced-in plots have not made up

Table VII.

Pruning weights (lb. new wood per tree).

Treatment.	Mixture.	Sub-plot.	Period I.		Period II.					
			Manures.	1941.	*New Manures.*	1943.	1944.	1945.	1946.	TOTAL 1943-6.
I. Swards frequently cut.	A	V	*0*	0·6	*10N*	0·5	0·9	2·2	2·7	6·3
		W	*2½N*	0·5	*7½N*	0·4	0·5	1·9	2·2	5·0
		X	*5N*	1·4	*5N*	1·4	1·3	3·4	3·9	10·1
		Y	*4P*	0·7	*4P*	0·4	0·4	1·2	2·0	4·0
		Z	*2½N* *4P*	0·8	*7½N* *4P*	1·0	1·1	3·0	3·5	8·5
		Mean		**0·8**		**0·7**	**0·8**	**2·3**	**2·9**	**6·8**
	B	V	*0*	1·2	*10N*	1·4	1·8	3·6	4·1	10·9
		W	*2½N*	1·3	*7½N*	1·4	1·5	3·4	4·2	10·5
		X	*5N*	1·3	*5N*	1·6	1·6	3·8	4·6	11·7
		Y	*4P*	1·0	*4P*	0·7	0·7	1·5	2·5	5·4
		Z	*2½N* *4P*	0·9	*7½N* *4P*	1·2	1·5	3·6	4·5	10·7
		Mean		**1·1**		**1·2**	**1·4**	**3·2**	**4·0**	**9·8**
	C	W	*2½N*	2·1	*7½N*	1·1	1·6	3·2	4·0	9·9
		V→X	*0*	2·6	*5N*	1·1	1·2	2·8	3·4	8·4
		Z	*2½N* *4P*	1·8	*7½N* *4P*	0·9	0·9	2·3	2·8	6·9
		Mean		**2·1**		**1·0**	**1·2**	**2·8**	**3·4**	**8·4**
II. Swards infrequently cut.	A	V	*0*	0·7	All cultivated, with uniform *N*.	0·9	2·8	3·1	2·6	9·4
		W	*2½N*	0·3		0·5	2·0	2·4	1·9	6·7
		X	*5N*	0·2		0·6	1·5	2·0	1·6	5·7
		Y	*4P*	0·6		0·6	2·4	2·4	2·6	8·0
		Z	*2½N* *4P*	0·6		0·4	1·9	2·9	2·3	7·6
		Mean		**0·5**		**0·6**	**2·1**	**2·6**	**2·2**	**7·5**
	B	V	*0*	0·8		1·3	3·2	3·7	2·8	11·0
		W	*2½N*	0·8		1·3	2·9	3·1	2·8	10·1
		X	*5N*	0·6		1·8	3·5	3·1	2·6	11·0
		Y	*4P*	0·5		1·2	2·2	2·9	2·2	8·5
		Z	*2½N* *4P*	0·6		1·0	2·5	2·8	2·0	8·3
		Mean		**0·7**		**1·3**	**2·9**	**3·1**	**2·5**	**9·8**
III. Cultivated				**4·2**		**2·1**	**2·5**	**3·7**	**3·5**	**11·9**

Apparent errors in the totals are due to suppression of decimals.

for the check during growth of the sward. After 7 seasons in grass, however, many of the trees in cutting treatment I were showing in 1946, new wood production greater than in the cultivated plots. Treatment Y, with no added nitrogen, gave noticeably poor new growth. None of these results has, however, been subjected to statistical analysis.

LEAVES.

The appearance of the leaves often gives an indication of the general nutritional condition of the tree and, besides periodical notes on appearance, detailed visual records of leaf colour were made in the summer. The time of autumn defoliation was recorded and this gave an indication both of rate of maturing of leaf, and of length of the period of leaf activity. Leaf scorch was also recorded, though it was not severe and relatively small areas of the leaf were affected.

Leaf colour. Leaf colour was estimated on each tree separately by visual inspection. In each of the years 1942 to 1945, a system of categorization was used in which 1 = yellow-green and 4 = dark green, an attempt being made to retain the same standard from year to year by reference to the Horticultural Colour Chart (British Colour Council and Royal Horticultural Society, 1938, 1941). In 1941, 5 categories were used. The mean values are shown in Table VIII.

No conspicuous differences in leaf colour were observed during the first season after sowing (1940) but, in 1941, nitrogen starvation symptoms appeared on the foliage of the grassed trees in early June on all plots. These symptoms became worse in 1942, when the trees with no nitrogen (treatments V and Y) had small yellow-green leaves, while an improved colour was shown with $2\frac{1}{2}N$ (W) and still greater improvement with $5N$ (X). Colour was better on mixture B (containing clover) than on mixture A, but frequency of cutting appeared to have no effect.

A quick response to nitrogen dressings was shown by the leaf colour of manurial treatment V, in 1943, when $10N$ was applied to plots which had received no nitrogen since the start of the trial. Two months after fertilizer application, the leaves assumed a dark green colour similar to that of the control trees and this similarity in colour to that under cultivation was maintained in the following seasons. Of the other plots remaining in grass after 1943, none, not even treatment W, gave as dark a leaf colour. It seems that, under those conditions, $10N$ was required to maintain the nitrogen status of grassed trees and that $7\frac{1}{2}N$ was not quite enough to give parity with conditions of cultivation. Further, on the grass plots, it is possible to deduce from the data a general improvement in colour in 1945 and this may be attributed to a release of nitrogen from the accumulated plant residues, but such a conclusion is dependent on maintaining the same colour standards in recording, to within half a category, from season to season.

Turning now to treatment II, complete restoration of the deep green colour followed the discing-in of the grass in 1943, indicating a rapid release of nitrogen from the decomposing organic matter. Continuing in 1944 and 1945 under the same conditions as the cultivated plots, it was to be expected that this colour would be maintained.

Comparison of W and Z treatments throughout shows that phosphate had no darkening effect on the leaf colour, which presumably depended mainly on the amount of available nitrogen.

TABLE VIII.

Leaf colour (higher numbers indicate darker leaves).

Treatment.	Mixture.	Sub-plot.	Period I.			Period II.				
			Manures.	1941.	1942.	*New Manures.*	1943.	1944.	1945.	MEAN 1943-45.
I. Swards frequently cut.	A	V	*0*	2·4	1·1	*10N*	3·4	3·7	4·0	3·7
		W	*2½N*	2·3	1·7	*7½N*	3·0	3·0	3·8	3·3
		X	*5N*	2·6	2·9	*5N*	3·2	2·8	3·2	3·1
		Y	*4P*		1·4	*4P*	2·5	2·3	2·9	2·6
		Z	*2½N 4P*		1·8	*7½N 4P*	3·3	3·4	3·6	3·4
		Mean		**2·4**	**1·8**		**3·1**	**3·0**	**3·5**	**3·2**
	B	V	*0*	3·1	2·2	*10N*	3·8	4·0	3·9	3·9
		W	*2½N*	4·1	2·6	*7½N*	3·4	3·6	3·8	3·6
		X	*5N*	3·6	2·8	*5N*	3·3	3·3	3·3	3·3
		Y	*4P*		2·3	*4P*	2·8	2·4	3·0	2·7
		Z	*2½N 4P*		2·6	*7½N 4P*	3·7	3·7	3·8	3·7
		Mean		**3·6**	**2·5**		**3·4**	**3·4**	**3·6**	**3·5**
	C	W	*2½N*		2·2	*7½N*	3·5	3·5	4·0	3·7
		V → X	*0*		2·4	*5N*	3·0	3·0	3·8	3·3
		Z	*2½N 4P*		2·5	*7½N 4P*	3·5	3·5	4·0	3·7
		Mean			**2·4**		**3·3**	**3·3**	**3·9**	**3·5**
II. Swards infrequently cut.	A	V	*0*	2·8	1·4	All cultivated, with uniform N.	4·0	4·0	4·0	4·0
		W	*2½N*	2·5	1·5		4·0	4·0	4·0	4·0
		X	*5N*	2·7	2·0		3·9	4·0	4·0	4·0
		Y	*4P*		1·4		3·9	4·0	4·0	4·0
		Z	*2½N 4P*		1·8		4·0	4·0	4·0	4·0
		Mean		**2·7**	**1·6**		**4·0**	**4·0**	**4·0**	**4·0**
	B	V	*0*	4·0	2·4		4·0	4·0	4·0	4·0
		W	*2½N*	4·2	2·3		4·0	4·0	4·0	4·0
		X	*5N*	3·9	2·9		4·0	4·0	4·0	4·0
		Y	*4P*		2·4		4·0	4·0	4·0	4·0
		Z	*2½N 4P*		2·6		4·0	4·0	3·9	4·0
		Mean		**4·0**	**2·5**		**4·0**	**4·0**	**4·0**	**4·0**
III. Cultivated				**4·6**	**4·0**		**3·9**	**4·0**	**3·9**	**3·9**

Autumn defoliation. Autumn defoliation was estimated by recording the state of defoliation of each tree on a given day in categories such that 0 = no defoliation, 10 = 100 per cent. defoliation, judged visually each year on a date when a good range was observable. The mean percentages are summarized in Table IX which shows that, in many cases, earliness of defoliation follows the same pattern as shortage of nitrogen, as found by Hoblyn and Bane (1934). For instance, in 1942 the trees with the yellowest leaf colour shed their leaves earliest (Tables VIII and IX). The differences between extremes were very striking, being about a month in some cases.

In later years the correlation between yellow leaf colour and earliness of defoliation is reversed in some cases. In 1943 and 1946 the plots remaining in sward retained their leaves considerably longer than did the cultivated strip. This is apparent even in sub-plots Y which received no nitrogen (Table IX). Thus it appears that, while nitrogen has a powerful effect in keeping the leaves green and functioning longer, some other factor, associated with swards, also helps in leaf retention. It would be interesting to test whether this may be a growth substance secreted by soil organisms which, as already mentioned (p. 243), are stimulated by the presence of organic matter.

Leaf scorches. Worcester Pearmain is a variety which does not readily show leaf scorches caused by mineral deficiencies and no leaf scorches were observed in this trial until the record crop year 1945. In that year some marginal scorch was observed and was probably due to the heavy demands of the fruit for potash ; some symptoms attributed to lack of magnesium and manganese were also noted. The extent of these symptoms was small (up to 10 per cent. of the leaves in the case of marginal scorch), but they were more prevalent on the cultivated plots than on the other treatments. Neither grass nor cultivated plots received any potash fertilizers during the trial and it is possible that the grass in some way makes available for the tree the potash reserves in the soil. It would scarcely be justifiable to draw a definite conclusion to this effect based on these observations alone, but it is of interest to note that a similar result was obtained with short-term leys in respect of magnesium (Rogers and Raptopoulos, 1945). Wallace also noted a lessening of leaf scorch symptoms when orchards were grassed down. He attributed this (1928) to a lowering of the nitrogen/potash ratio, but later data for a plot at East Malling (Wallace and Proebsting, 1933) suggested that grassing down actually increased the amount of available potash in the soil.

TIME OF BLOSSOMING.

Lack of available nitrogen has been associated with a delay in the opening of the blossoms. Retardation of blossoming associated with growing cover crops was described in a previous paper of this series (Rogers and Raptopoulos, 1945), in which it was pointed out that the effect might be accounted for by a relative deficiency of nitrogen. However, detailed visual records of amount of blossom open at a given date have failed to show any striking differences in blossom period of the trees, between the different swards, manures, or cultural treatments in this experiment.

FRUIT.

The crop was recorded for each tree, picked fruit and dropped fruit being considered separately and then combined to give the total crop record summarized in Table X.

TABLE IX.

Autumn defoliation (percentage defoliation at a given date).

Treatment.	Mixture.	Sub-plot.	Period I.		Period II.				
			Manures.	1942.	*New Manures.*	1943.	1944.	1945.	1946.
I. Swards frequently cut.	A	V	*0*	93	*10N*	58	56	83	20
		W	*2½N*	88	*7½N*	68	62	82	21
		X	*5N*	73	*5N*	78	77	88	24
		Y	*4P*	80	*4P*	73	79	76	26
		Z	*2½N* *4P*	79	*7½N* *4P*	70	63	82	22
		Mean		**83**		**69**	**67**	**82**	**23**
	B	V	*0*	57	*10N*	51	38	87	19
		W	*2½N*	58	*7½N*	53	48	83	24
		X	*5N*	48	*5N*	66	61	89	23
		Y	*4P*	61	*4P*	63	68	79	20
		Z	*2½N* *4P*	56	*7½N* *4P*	65	49	86	13
		Mean		**56**		**60**	**53**	**85**	**20**
	C	W	*2½N*	77	*7½N*	65	52	72	20
		V → X	*0*	78	*5N*	81	71	73	33
		Z	*2½N* *4P*	73	*7½N* *4P*	52	50	77	17
		Mean		**76**		**66**	**58**	**74**	**23**
II. Swards infrequently cut.	A	V	*0*	97	All cultivated, with uniform N.	57	55	77	31
		W	*2½N*	99		63	56	72	24
		X	*5N*	86		61	57	74	29
		Y	*4P*	97		56	60	80	23
		Z	*2½N* *4P*	98		69	61	79	34
		Mean		**95**		**61**	**58**	**76**	**28**
	B	V	*0*	50		61	56	74	31
		W	*2½N*	58		57	54	74	38
		X	*5N*	40		71	55	80	28
		Y	*4P*	46		64	60	70	31
		Z	*2½N* *4P*	48		60	50	67	22
		Mean		**48**		**63**	**55**	**73**	**30**
III. Cultivated				**87**		**87**	**63**	**82**	**37**

Total crop (lb. per tree).

Treatment.	Mixture.	Sub-plot.	Period I.					Period II.						GRAND TOTAL.
			Manures.	1940.	1941.	1942.	TOTAL.	*New Manures.*	1943.	1944.	1945.	1946.	TOTAL.	
I. Swards frequently cut.	A	V	0	55	76	25	155	10N	98	13	140	149	399	554
		W	2½N	53	60	26	139	7½N	76	14	127	135	351	490
		X	5N	66	80	60	206	5N	120	23	152	169	464	670
		Y	4P	50	71	26	146	4P	99	10	106	160	375	521
		Z	2½N 4P	54	65	27	146	7½N 4P	96	12	129	144	382	528
		Mean		**56**	**70**	**33**	**159**		**98**	**14**	**131**	**151**	**394**	**553**
	B	V	0	53	71	48	172	10N	135	12	127	146	421	593
		W	2½N	59	69	80	207	7½N	137	17	143	148	444	651
		X	5N	58	80	91	228	5N	137	27	153	174	491	719
		Y	4P	52	57	31	139	4P	89	16	112	139	356	495
		Z	2½N 4P	47	56	51	155	7½N 4P	109	18	123	137	387	542
		Mean		**54**	**66**	**60**	**180**		**121**	**18**	**132**	**149**	**420**	**600**
	C	W	2½N	64	69	76	209	7½N	128	37	175	160	500	709
		V → X	0	55	58	76	188	5N	104	55	178	156	493	681
		Z	2½N 4P	64	58	85	207	7½N 4P	117	22	157	144	440	647
		Mean		**61**	**61**	**79**	**201**		**116**	**38**	**170**	**153**	**478**	**679**
II. Swards infrequently cut.	A	V	0	54	60	32	146	All cultivated, with uniform N.	147	19	162	125	453	599
		W	2½N	55	64	21	140		130	19	190	132	472	611
		X	5N	50	54	39	143		137	17	159	120	434	577
		Y	4P	56	57	19	132		154	22	169	130	474	606
		Z	2½N 4P	49	54	23	125		139	9	150	116	413	538
		Mean		**53**	**58**	**27**	**137**		**141**	**17**	**166**	**125**	**449**	**586**
	B	V	0	47	63	50	160		162	16	171	149	498	659
		W	2½N	46	62	59	167		159	11	171	118	459	626
		X	5N	45	52	55	152		142	11	160	129	442	594
		Y	4P	43	61	43	147		154	9	162	133	458	605
		Z	2½N 4P	48	66	57	171		147	9	178	147	482	653
		Mean		**46**	**61**	**53**	**159**		**153**	**11**	**168**	**135**	**468**	**627**
III. Cultivated				**53**	**72**	**107**	**232**		**117**	**31**	**135**	**153**	**436**	**668**

Apparent errors in the totals are due to suppression of decimals.

Until 1945, all the fruit was weighed to 0·1 lb., but in 1945 and 1946 the heavy crop and shortage of labour necessitated the use of a quicker recording method ; in those years all records were made in number of bushel boxes (to nearest ¼ box) and then converted to pounds by the use of the mean weight of a number of boxes.

Considerable differences were noted, and are discussed in detail below, in the relative proportion of picked and dropped fruit between treatments. The picked fruit, representing all the more readily marketable apples, is given in Table XI, while the data for percentage of "drops" appear in Table XII. The 1940 entries in Table XI are identical with those in Table X, because no record of dropped fruit was made in that year and the assumption has been made, for that year only, that the picked fruit record was a reasonable indication of the total crop.

Fruit size was estimated by weighing known numbers of apples from each treatment. The colour of the fruit was recorded on the tree by the visual method described in the first paper of this series (Rogers and Raptopoulos, 1945) in which both intensity of ground colour and proportion of surface reddening were estimated. In this investigation no striking differences were observed in intensity of ground colour, so the data are not given, but the colour data show considerable differences in proportion of surface reddening (Table XIII).

Other records which were made were on fruit firmness, in 1945, and on amount of apple scab (*Venturia inaequalis*) in 1941 and 1942.

In the crop records, in order to supplement the data of the single cultivated strip, records were made of a further set of cultivated trees at the south end of the plot. It was found that these data confirmed those of the experimental strip and thus gave additional support to the results obtained.

In general it was found that those swards and treatments which checked the tree very severely also reduced the crop. Treatments which had relatively little checking effect had little or no effect on crop. On practically all sward treatments great improvements in fruit quality and market value were obtained, both in improved fruit colour and in lessened premature dropping of fruit.

The various crop records will be considered in detail, in the two periods of the experiment separately.

PERIOD I (1940-42).

Changes in the effects of the different treatments occurred from year to year in this period.

Total crop. In the year of sowing of the cover crops (1940), when no manures were applied, it will be seen (Table X) that the yields of plots V to Z under similar treatments varied above and below that of the cultivated trees. This shows the natural variability of the material, and none of the differences is significant. In 1941 the swards began to have a checking effect on cropping and in 1942 this had become severe, especially in plots of mixture A, plots infrequently cut, and sub-plots receiving little nitrogen (Table X). The variability was so great, however, that the large difference between the 1942 crop under frequently cut grass and that under cultivation was not significant at $P = 0{\cdot}05$:

	lb. per tree.
Cutting treatment II (infrequent)	40
Cutting treatment I (frequent)	46
Cultivation III	107

Treatment.	Mixture.	Sub-plot.	Period I.					Period II.						GRAND TOTAL.
			Manures.	1940.	1941.	1942.	TOTAL.	*New Manures.*	1943.	1944.	1945.	1946.	TOTAL.	
I. Swards frequently cut.	A	V	*0*	55	69	23	147	*10N*	88	10	95	88	281	428
		W	*2½N*	53	55	25	133	*7½N*	70	12	93	96	271	404
		X	*5N*	66	71	51	188	*5N*	109	20	108	126	362	550
		Y	*4P*	50	63	23	136	*4P*	94	9	82	132	317	452
		Z	*2½N 4P*	54	58	24	137	*7½N 4P*	86	11	87	100	283	420
		Mean		**56**	**63**	**29**	**148**		**89**	**12**	**93**	**108**	**303**	**451**
	B	V	*0*	53	62	43	159	*10N*	121	10	91	84	305	464
		W	*2½N*	59	60	74	192	*7½N*	124	14	101	100	339	531
		X	*5N*	58	69	77	203	*5N*	123	20	117	116	376	579
		Y	*4P*	52	51	27	130	*4P*	84	14	93	112	303	433
		Z	*2½N 4P*	47	47	41	135	*7½N 4P*	95	14	90	99	298	433
		Mean		**54**	**58**	**52**	**164**		**109**	**14**	**98**	**102**	**324**	**488**
	C	W	*2½N*	64	56	68	189	*7½N*	120	32	98	143	393	581
		V → X	*0*	55	44	65	164	*5N*	96	47	123	137	402	566
		Z	*2½N 4P*	64	49	76	189	*7½N 4P*	109	18	101	133	360	549
		Mean		**61**	**50**	**70**	**180**		**108**	**32**	**107**	**138**	**385**	**565**
II. Swards infrequently cut.	A	V	*0*	54	51	30	136	All cultivated, with uniform N.	130	15	123	84	351	487
		W	*2½N*	55	59	20	133		114	14	145	89	363	495
		X	*5N*	50	51	37	137		122	14	116	84	336	473
		Y	*4P*	56	47	18	121		140	17	135	85	377	498
		Z	*2½N 4P*	49	46	21	116		124	7	112	70	313	429
		Mean		**53**	**51**	**25**	**128**		**126**	**13**	**126**	**82**	**348**	**477**
	B	V	*0*	47	55	44	147		136	11	120	86	354	501
		W	*2½N*	46	53	54	153		140	9	128	65	343	496
		X	*5N*	45	48	48	141		118	8	112	76	315	456
		Y	*4P*	43	53	39	135		132	6	119	78	335	470
		Z	*2½N 4P*	48	58	51	156		126	7	122	85	340	497
		Mean		**46**	**53**	**47**	**146**		**131**	**8**	**120**	**78**	**337**	**484**
III. Cultivated				**53**	**60**	**74**	**187**		**97**	**23**	**78**	**67**	**264**	**451**

Apparent errors in the totals are due to suppression of decimals.

TABLE XII.

Percentage of drops.

Treatment.	Mixture.	Sub-plot.	Period I.				Period II.					
			Manures.	1941.	1942.	1941-42 inclusive.	*New Manures.*	1943.	1944.	1945.	1946.	1943-46 inclusive.
I. Swards frequently cut.	A	V	*o*	9	7	8	*10N*	10	17	32	41	30
		W	*2½N*	7	6	7	*7½N*	8	10	27	29	23
		X	*5N*	11	15	13	*5N*	10	14	29	26	22
		Y	*4P*	12	10	11	*4P*	6	6	23	18	16
		Z	*2½N* } *4P* }	10	11	10	*7½N* } *4P* }	11	13	33	31	26
		Mean		**10**	**10**	**10**		**9**	**12**	**29**	**29**	**23**
	B	V	*o*	12	10	11	*10N*	11	20	29	43	28
		W	*2½N*	13	8	10	*7½N*	10	18	29	33	24
		X	*5N*	14	15	15	*5N*	10	27	24	34	24
		Y	*4P*	10	10	10	*4P*	6	12	17	19	15
		Z	*2½N* } *4P* }	16	20	18	*7½N* } *4P* }	13	21	27	28	23
		Mean		**13**	**13**	**13**		**10**	**20**	**25**	**31**	**23**
	C	W	*2½N*	18	11	14	*7½N*	6	15	44	11	22
		V → X	*o*	23	15	19	*5N*	8	15	31	12	18
		Z	*2½N* } *4P* }	16	10	12	*7½N* } *4P* }	7	21	36	8	18
		Mean		**19**	**12**	**14**		**7**	**17**	**37**	**10**	**19**
II. Swards infrequently cut.	A	V	*o*	14	6	11	All cultivated, with uniform N.	12	21	24	33	22
		W	*2½N*	8	8	8		12	26	24	33	23
		X	*5N*	7	6	6		11	22	27	30	23
		Y	*4P*	18	6	15		9	23	20	35	21
		Z	*2½N* } *4P* }	14	6	12		11	18	25	40	24
		Mean		**12**	**6**	**11**		**11**	**22**	**24**	**34**	**22**
	B	V	*o*	12	13	12		16	29	30	42	29
		W	*2½N*	14	9	12		12	18	25	45	25
		X	*5N*	8	13	10		17	26	30	41	29
		Y	*4P*	13	9	11		14	32	27	42	27
		Z	*2½N* } *4P* }	13	11	12		14	22	32	42	29
		Mean		**12**	**11**	**11**		**15**	**25**	**29**	**42**	**28**
III. Cultivated				**17**	**30**	**25**		**17**	**28**	**43**	**56**	**39**

Fruit colour (percentage of surface coloured red).

Treatment.	Mixture.	Sub-plot.	Period I.				Period II.				
			Manures.	1941.	1942.	MEAN.	*New Manures.*	1943.	1944.	1945.	MEAN.
I. Swards frequently cut.	A	V	0	79	74	77	10N	53	44	63	53
		W	2½N	69	71	70	7½N	61	51	68	60
		X	5N	67	64	66	5N	64	55	69	63
		Y	4P		70		4P	77	68	80	75
		Z	2½N 4P		69		7½N 4P	57	48	62	56
		Mean		**72**	**70**	**71**		**62**	**53**	**68**	**61**
	B	V	0	66	65	66	10N	48	38	58	48
		W	2½N	60	65	63	7½N	49	38	66	51
		X	5N	60	60	60	5N	55	45	68	56
		Y	4P		67		4P	65	59	80	68
		Z	2½N 4P		62		7½N 4P	51	42	66	53
		Mean		**62**	**64**	**63**		**54**	**44**	**68**	**55**
	C	W	2½N				7½N	55	40	61	52
		V → X	0				5N	59	43	68	57
		Z	2½N 4P				7½N 4P	58	43	61	54
		Mean						**57**	**42**	**63**	**54**
II. Swards infrequently cut.	A	V	0	70	72	71	All cultivated, with uniform N.	41	33	51	42
		W	2½N	77	77	77		44	39	66	50
		X	5N	70	73	72		44	40	62	49
		Y	4P		78			44	37	58	46
		Z	2½N 4P		77			43	35	61	46
		Mean		**72**	**75**	**73**		**43**	**37**	**60**	**47**
	B	V	0	71	71	71		44	38	56	46
		W	2½N	70	70	70		43	34	62	46
		X	5N	72	65	69		43	35	59	46
		Y	4P		68			46	38	59	48
		Z	2½N 4P		63			46	38	62	49
		Mean		**71**	**67**	**69**		**44**	**37**	**60**	**47**
III. Cultivated				**29**	**32**	**31**		**43**	**40**	**50**	**44**

The difference between treatments II and III was just significant. An attempt was made to eliminate the variation due to initial differences between the trees by adopting the method of covariance, using the area of cross-section of the trunk in 1939 as an indication of the cropping capabilities of the trees at the start of the investigation. When this was done, however, no further significant difference appeared, indeed even the difference between treatments II and III then ceased to be significant.

The corresponding treatment means for the period as a whole appear as follows :

	lb. per tree.
Cutting treatment II (infrequent)	148
Cutting treatment I (frequent)	169
Cultivation III	232

Both cutting treatments gave a significantly lower crop than did cultivation. This difference ceased to be significant when the data were corrected by 1939 cross-sectional area of trunk, but was significant when the 1934-39 total crop was used as a basis for correction. In no case was the difference between cutting treatments significant. Mixture B, however, gave a significantly higher yield than did mixture A, considering the data corrected by the 1939 area of cross-section of the trunk :

	lb. per tree.
Mixture A	149
Mixture B	169

Mixture C could not be included in the analysis, but its yields compare very favourably with those of comparable treatments in mixtures A and B (Table X). A comparison of swards A and B, and of frequent and infrequent cutting, is given in Fig. 7.

The total crop in the years 1941-42 for the sub-plots receiving different manurial treatments in those years was also analysed. The data, corrected by the 1939 area of trunk cross-section, appear thus (lb. per tree) :

	Cutting treatments.	
Manures	*I*	*II*
V (0)	111	102
W $(2\frac{1}{2}N)$	115	105
X $(5N)$	147	107

On plots receiving frequent cutting (I), treatment X $(5N)$ gave significantly higher crops than did the other treatments. There were no significant differences between manurial treatments on plots receiving cutting treatment II. Thus, while an application of 2½ cwt. per acre of sulphate of ammonia had no significant effect, a further 2½ cwt. resulted in a significant crop increase when the grass was mown frequently. On the infrequently mown plots, apparently even 5 cwt. of sulphate of ammonia was all used up by the increased growth of grass.

In fact, for the 1940-42 period as a whole, the only grass sub-plots which maintained a total yield comparable with that of the cultivated treatment were those of mixture B, frequently cut, and receiving 5 cwt. per acre of sulphate of ammonia (Table X and Fig. 6) and, to a lesser extent, those of mixture C receiving nitrogen.

As superphosphate was not applied till 1942, the treatments Y $(4P)$ and Z $(4P+2\frac{1}{2}N)$ were analysed for that year only, compared with V (*nil*), but no effects were found significant. The effect of N and P dressings on sub-plots of sward B are shown graphically in Fig. 6.

Fruit dropping. A striking and important contrast was found in the amount of prematurely dropped fruit from 1941 onwards. Table XII shows that, both in 1941 and 1942, the percentage of dropped fruit was much less on the grass plots than on the cultivated ones. In 1942 many of the grass plots showed less than 10 per cent. "drops" as compared with 30 per cent. under cultivation conditions. As the picked crop is, of course, of greater market value than dropped fruits, this effect is of great commercial importance; further, owing to the reduced damage to the fruit when falling on a grass sward, a far greater proportion of the dropped fruit is marketable (as "drops") on grass than on the cultivated plots.

In picked crop the grassed plots thus compare more favourably with the cultivated plots than they did in total crop, as shown in Table XI. This shows that, on the frequently cut swards, more fruit was picked from sub-plots W and X on mixture B, X on mixture A, and W and Z on mixture C, than from the cultivated plot during the period. The middle and bottom parts of Fig. 6 show the same effect.

Work by Southwick (1940) has suggested that reduced drop may be connected with reduced nitrogen supply. This may be the case here, for in 1941 and 1942 the grass was presumably competing fairly severely with the trees for nitrogen but, as is discussed later, other factors also seem to be involved.

Fruit colour. The effects of the swards on fruit colour were very striking. Table XIII shows that in 1941 and 1942 the fruits on grass plots had more than twice as much red colour as those on the cultivated plots. This effect, previously noted by Hoblyn and Bane (1934), is probably also related to the nitrogen status of the plots (Gourley and Howlett, 1941, p. 176). The plots receiving least added nitrogen and those in which competition from the grasses was most severe had the highest coloured fruit but addition of 5 cwt. of sulphate of ammonia to the swards did not appreciably lessen the improvement in fruit colour. It can be seen that plots of mixture A had rather more highly coloured fruits than those of mixture B.

This big improvement in colour of the apples, gained by the use of grass, is of special importance in coloured varieties such as Worcester Pearmain.

Fruit size. The data for fruit size did not show any large or consistent contrasts, so the figures are not given in detail here. In 1941, the fruit from most grassed plots averaged 13 lb. per 100 fruits, compared with 15 lb. for those from cultivated plots, but in 1942 the order was reversed and fruits from grassed plots averaged 16 or 17 lb. per 100 fruits, while those from cultivated plots remained at 15 lb. Thus any competing effect of the grasses has evidently affected number rather than size of fruits. The manures showed no appreciable effects on fruit size in this case.

Scab. In 1941 and in 1942 the fruit from this trial was graded by Mr. M. H. Moore for severity of incidence of Scab. The only significant difference between treatments in 1941 was that the fruit on the grass plots mown infrequently (II) showed less Scab than those on the frequently mown (I) plots. This effect, however, was not confirmed in 1942, when no differences were found to be significant. Moore (1936) had found previously that grassing-down decreased the tendency to Scab, but no such difference appeared in the present investigation.

General. Summing up the crop records for 1940-42, grassing-down enormously improved fruit colour and reduced fruit drop but tended to reduce the total crop where competition was most severe, i.e. in the infrequently cut swards, in mixture A, and in plots receiving less than 5 cwt. of sulphate of ammonia. Plots under mixture

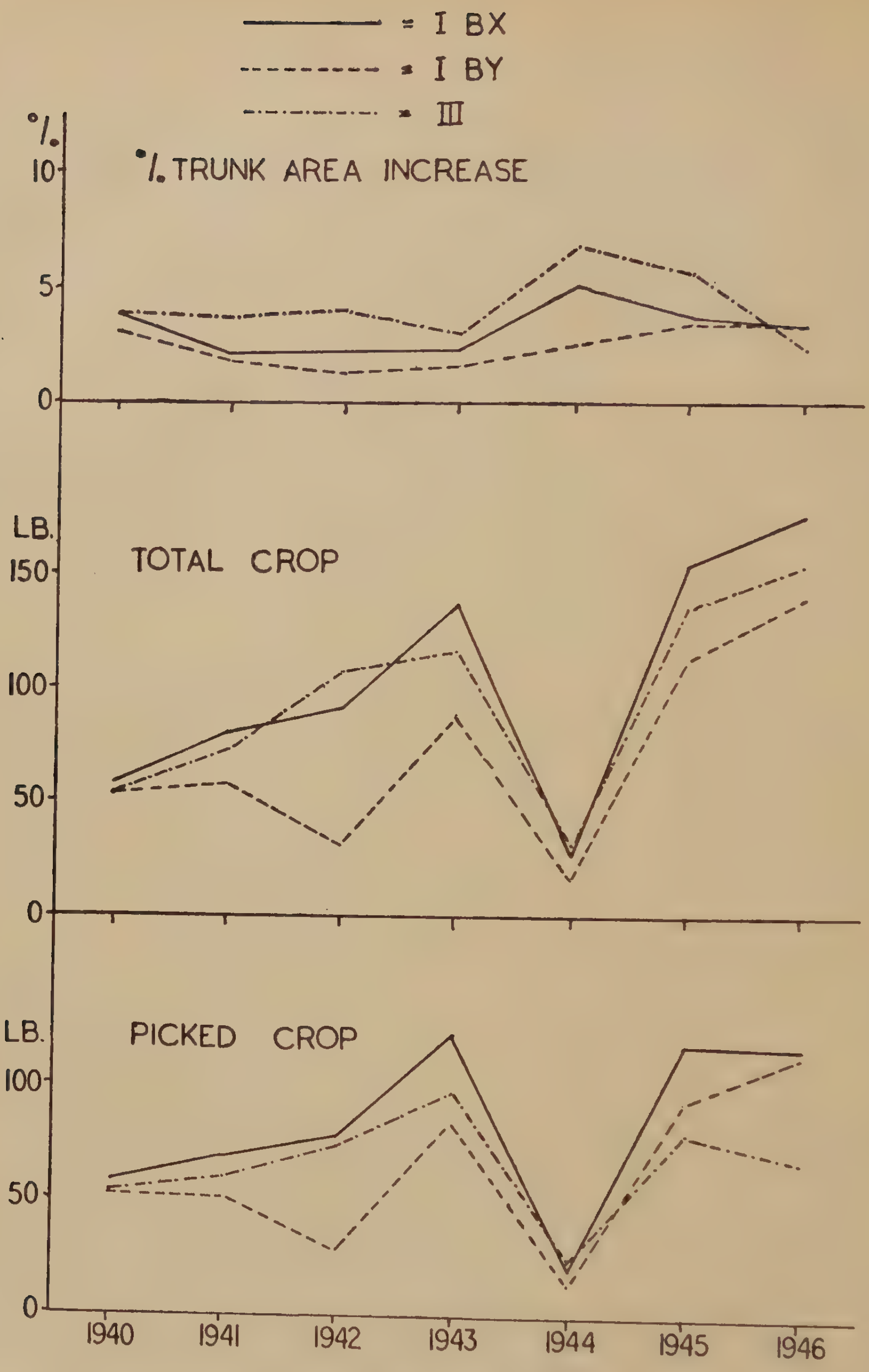


FIG. 6.

Performance of Worcester Pearmain under swards with different manuring, compared with cultivation.

I BX = frequently cut sward of mixture B receiving 5 cwt. per acre sulphate of ammonia per year (also in Fig. 7).

I BY = frequently cut sward of mixture B receiving no nitrogen but 4 cwt. per acre superphosphate per year.

III = cultivated, with autumn weeds, receiving no manures in 1940-44, then 3 cwt. per acre sulphate of ammonia in 1945 and again in 1946.

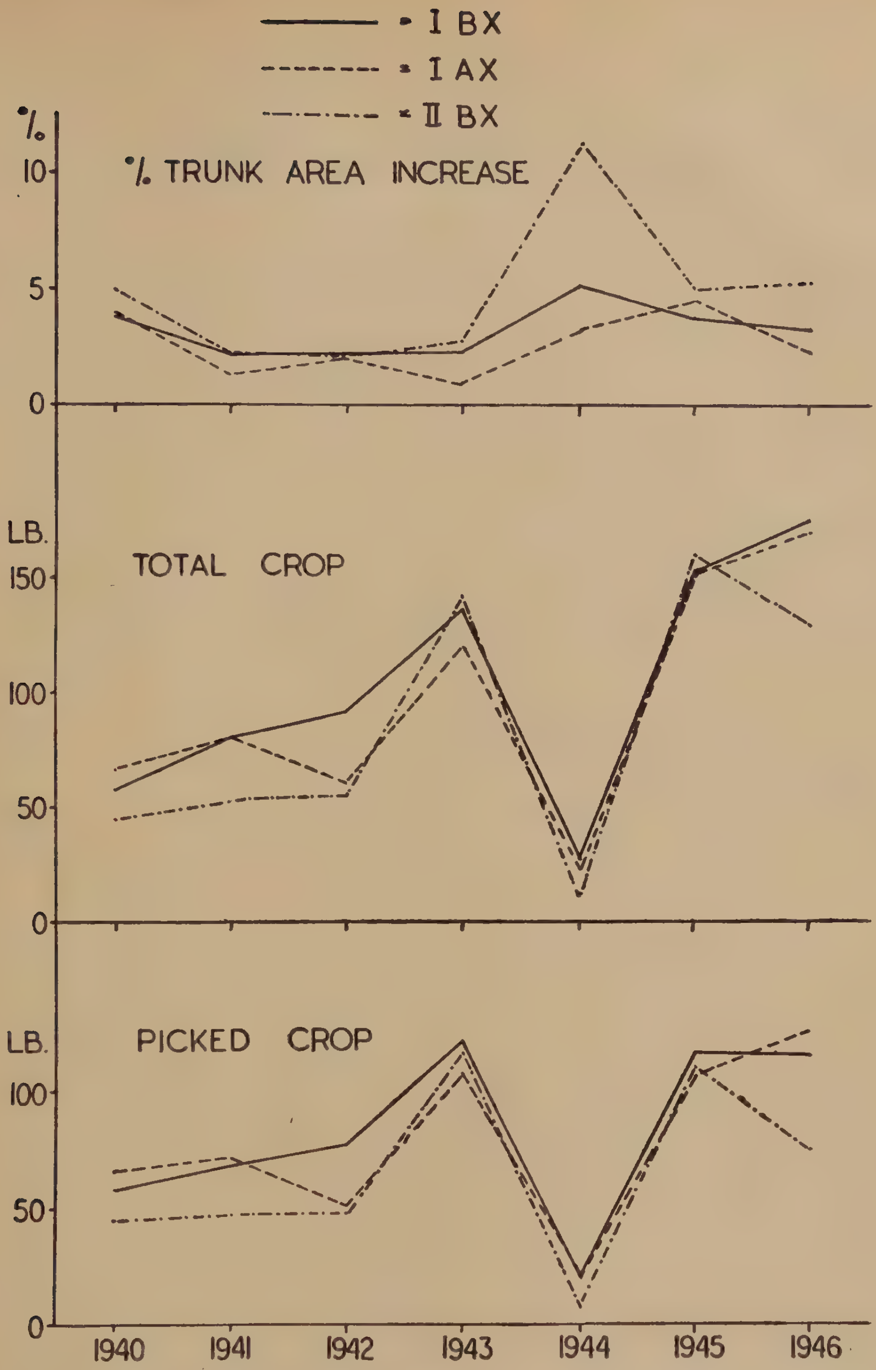


FIG. 7.

Performance of Worcester Pearmain under two different swards frequently cut, and under an infrequently cut ley.

I BX = frequently cut sward of mixture B receiving 5 cwt. per acre sulphate of ammonia per year (also in Fig. 6.).

I AX = frequently cut sward of mixture A receiving 5 cwt. per acre sulphate of ammonia per year.

II BX = infrequently cut sward (ley) of mixture B receiving 5 cwt. per acre sulphate of ammonia per year 1941-42, turned-in in 1943 with 2 cwt. per acre sulphate of ammonia, then cultivated, receiving no manures in 1944, then 3 cwt. per acre sulphate of ammonia in 1945 and again in 1946.

B, cut frequently and manured with 5 cwt. of sulphate of ammonia showed improved fruit colour and lessened drop without reduction in amount of crop.

PERIOD II (1943-46).

As already stated, the grass on plots receiving treatment II was disced-in in 1943 and thereafter all such plots received uniform manurial treatment identical with that applied on the cultivated strip (treatment III). At the same time some of the manurial treatments on the sub-plots of the continued sward plots (treatment I) were changed. Sub-plots X and Y continued to have the same manures, but sub-plots V, W and Z, which previously had small nitrogen dressings, now had large ones. The changes, together with the total crop figures, are shown in Table X.

Total crop. Considering first the grass plots, Table X shows that sward B retained its superiority over sward A in 1943, but in 1945 and 1946 the difference became negligible. This can be seen well in Fig. 7 which also shows clearly the great reduction of crop caused by frost in 1944 and the stimulus to growth which accompanied it. Differences between manurial treatments failed to reach statistical significance. The X sub-plots, especially on mixture B, still gave the highest figures. The sub-plots on sward C could not be included in an analysis but their crops were also very good.

The turned-in plots (treatment II) showed a great stimulus to cropping in 1943 and this stimulus continued in 1945 but the crop was lower than that of the cultivated plots in 1946. There seems little effect of difference in former swards or manurial dressings.

Comparing the main cultural treatments, the totals for the period appear as follows :

	lb. per tree.
Treatment I (frequently cut sward)	407
Treatment II (disced-in sward)	458
Cultivated III	436

In statistical analysis the crop data were covarianced on to a function of the 1942 girth and the difference between the corrected figures for treatments I and II just fails to be significant. There is little doubt, however, that the turned-in sward had a tendency to improve cropping as well as growth, at any rate up to 1945.

Fruit dropping. The marked reduction in fruit dropping continued on the grass plots, though there is an indication that the increased nitrogen dressings tended to decrease the effect (Table XII). The cultivated plots in this period gave a lower picked crop than did any separate combination of seeds mixture and manurial treatment (Table XI).

An interesting feature of the turned-in swards was that they also continued to exercise a favourable effect on fruit retention (Table XII). The contrast is not so great as under continued sward but, even so, the mean picked crop over this period on all turned-in sward plots was some 30 per cent. greater than that on the cultivated plots (Table XI).

The very heavy " drop " figures for 1946 are due to a gale at picking time.

Fruit colour. The fruit colour remained excellent on the grassed plots though, here again, the very heavy nitrogen dressings tended to reduce it to some extent. The plots receiving no nitrogen continued to show the highest fruit colour. In 1944, when the fruits were very few in all plots, the colour was relatively poor.

On the plots of turned-in sward, the fruit colour at once reverted to that of the cultivated plots. This also suggests that available nitrogen is one of the main factors involved.

Fruit size. The data for fruit size once more show practically no difference between continued swards, turned-in swards and clean cultivation throughout the period.

Fruit hardness. A study of the firmness of the flesh of apples from grass and cultivated plots was made in 1945. Readings were made of the pressure required to drive a cylindrical plunger of diameter 7 mm. to a depth of 12 mm. into the flesh (after removal of the peel). No significant differences were obtained.

General. The crop records of 1943-46 have provided evidence that the nitrogen deficiency which developed under certain treatments in the previous period was corrected by increased nitrogen dressings. Further, there was evidence that the swards, having " locked up " nitrogen for some seasons, were now beginning to " pay back ", releasing nitrogen available for the trees. Under these conditions, addition of 5 cwt. per acre of sulphate of ammonia was found to be sufficient, provided that the trees were not nitrogen deficient in the first place. The previous large differences between the effects of swards A and B almost disappeared in 1943-46, possibly due to changes in the composition of the swards.

BOTH PERIODS TOGETHER.

Crop figures for the whole of the first seven years of the trial are given at the right of Tables X and XI. In plots on which the treatments were changed during the period, these totals need to be interpreted with caution but some interesting comparisons are available.

The data for treatment II show the effect of a three-year ley of strong grass, infrequently cut, followed by four years cultivation. Over the seven seasons the total crop (Table X) was in every instance lower than that of the plots cultivated throughout. Evidently the initial check was too severe. On the other hand, because of the decreased severity of fruit dropping (Table XII) both during and after the life of the sward, this three years' ley and four years' cultivation gave, in nine cases out of ten, a higher amount of *picked* fruit than the cultivated strip.

On the continued grass plots, treatments X and Y which remained unchanged throughout the investigation, are of special interest. Treatment X, which received 5 cwt. per acre of sulphate of ammonia throughout, gave on mixture B the highest crop of all treatments. On mixture A also, its yield was the highest of its class ; this in spite of the fact that the *total* amount of nitrogen added in the seven years was less than in any other treatment except Y. Evidently lack of nitrogen in early years cannot easily be offset by increased dressings later.

Treatment Y, which received no nitrogen but 4 cwt. per acre of superphosphate each year, gave much smaller total crops than either treatment II or the cultivated strip. On the other hand, the percentage of fruit dropping was lower than on other treatments, so that the actual amount of picked fruit (Table XI) in the case of treatment Y on mixture A was about equal to that of the cultivated strip. The fruit colour also was higher on these plots than on all others. These advantages, however, are not sufficient to offset the disadvantages of too low nitrogen status in its effect on tree growth and cropping.

Mixture C, being outside the main trial, has not been given much attention in the discussion. The high yields, however, call for some comment. Whether total crop (Table X) or picked crop (Table XI) be considered, the yields on mixture C for each manurial treatment compare well with the highest (treatment X) on mixtures A and B. Comparison of W with Z, as in the case of mixtures A and B, shows the effect of superphosphate to be inappreciable. As no nitrogen was applied on treatment W and Z till 1942 and V → X received nothing till 1943, the performance is all the more remarkable and suggests that such weak grasses which cause only a very gentle check in early years, are worthy of a place in a full scale trial in future.

In general terms, the crop records suggest that the system of management likely to give most satisfactory results under similar conditions may be one involving a weak-growing sward, frequently mown, and given about 5 cwt. per acre of sulphate of ammonia per year.

DISCUSSION AND CONCLUSIONS.

The problem under investigation was how to produce organic material, for the benefit of the soil, by the relatively simple method of permanent swards or long-term leys in the orchard, without harming the trees. The present trial has shown that this can be done successfully but that success depends on choice of a suitable sward, fertilizer and cutting programme. Given these conditions, not only can large amounts of organic matter be produced but the trees can benefit considerably both in quality and amount of crop, under the soil and climatic conditions of East Malling. With unsuitable swards, cutting or fertilizer programme, a severe check to tree growth and cropping may be expected.

The results show, for conditions such as those tested, that vigorous grasses like cocksfoot must be avoided ; that mowing should be frequent (at least six times a year) and that, at least in the first three or four years, generous applications of nitrogen (about 5 cwt. or more per acre of sulphate of ammonia), must be applied in order to maintain satisfactory tree growth and cropping. Given these conditions the trees will not suffer and, further, the sod represents a store of fertility which can be cashed by turning-in, resulting at once in a striking acceleration of growth and cropping if required.

There are certain advantages to be gained by the continued use of a sward, for instance great improvement of fruit colour and lessening of pre-harvest drop. There is also some evidence both from this and other experiments, that certain mineral deficiencies may be lessened.

Hoblyn and Bane (1934) showed the harmful effects of grassing-down without added nitrogen and the deterioration in fruit quality resulting from addition of too much nitrogen. This experiment has emphasized the great importance of maintaining a correct nitrogen level. Too little will allow undue checking of the tree. Too much will lessen the benefits to fruit quality. Further work on this point, including varied time of application and possible deep placement, is desirable.

The use of weaker swards, including *Agrostis* and *Poa*, will also be worth further investigation.

From the point of view of direct effects on the soil, it is of interest to note that although large amounts of organic material are produced, the percentage of organic matter in the soil does not increase rapidly. This is because rapid decomposition is also stimulated. Thus the organic matter is not by any means all " banked " in a " deposit account ", but much of it remains in a " current account ". The resulting bigger " turnover ", however, seems definitely beneficial.

From the labour point of view, the use of swards has attractions also. Gang-mowing is as easy as cultivation, though in both there is still need for better implements which will go closer to the tree trunks without risk of damaging them. Mr. F. H. Villiers has estimated that in the East Malling orchards a single gangmowing operation, in two directions at right angles, costs 5s. 3½d. per acre, whereas cultivation in both directions cost 7s. 10d. per acre. Rather more mowings than cultivations are necessary per season, so there is probably little difference in the actual cost of the operations per season. But the presence of a sward makes the operations of pruning, spraying and picking much easier, and enables them to be done at the grower's convenience, without having to wait for the soil to dry, besides saving much time and labour.

The foregoing data have shown that different swards and systems of management vary in their checking or stimulating effect on tree growth. It is therefore clearly necessary, when choosing a seeds mixture and deciding on an appropriate system of management, to consider factors such as rootstock, type of tree, soil and climate (Rogers and Greenham, 1948).

SUMMARY.

1. In a trial of the use of swards in comparison with clean cultivation (with autumn weeds) in a plantation of 30-year-old Worcester Pearmain apples, three different grass mixtures, two frequencies of cutting and five different manurial treatments were tested.

The first seven years' results are reviewed.

2. The main ingredients of the mixtures were (A) cocksfoot, perennial ryegrass and wild white clover ; (B) perennial ryegrass, broad red and wild white clover ; (C) fine lawn grasses and wild white clover. All mixtures grew well under the trees and did not suffer from the orchard spraying routine.

3. The cutting treatments were (I) frequent (about six times a year) and (II) infrequent (about three times a year). The latter series was maintained for three years followed by four years' observation of the effect of the turned-in sward.

4. The manurial treatments investigated were various rates of application of sulphate of ammonia and a single rate of application of superphosphate with and without sulphate of ammonia. The treatments were changed on the swards after the first three years. The cultivated area and the disced-in swards received uniform dressings of sulphate of ammonia. No potash was applied at any time.

5. Large amounts of cut green material were produced by the swards, varying, according to the vigour of the sward and the treatment adopted, from about 2 tons to 10 tons per acre. The bulk of each year's production occurred by the end of May. By infrequent cutting, the yield was about twice that obtained by frequent cutting.

The yield of cut material responded to applications of sulphate of ammonia up to 7½ cwt./acre ; a further 2½ cwt./acre gave little extra response.

Mixture B exceeded mixture A in yield in early years when the broad red clover was growing strongly but in later years, when the broad red clover had died out and mixture A had become nearly pure cocksfoot, A exceeded B.

6. Clovers could be maintained only if nitrogen were withheld. The effect on the trees showed that the clover-produced nitrogen was, however, insufficient for satisfactory tree performance. Superphosphate stimulated the cover crop yield only in the presence of clovers.

7. The effect of the swards on spring air temperatures was to cause a reduction in minimum temperature, at a height of 5 ft. 6 in., of the order of 1° F. or less.

8. Despite the addition of large amounts of fresh material to the soil, the disced-in plots showed no increase in organic matter in the soil seven months after turning-in. The C/N ratios showed that decomposition had been rapid. The soil of these plots was, however, less compact than that of plots cultivated throughout.

9. In comparison with cultivated soil, the soil under grass was found to dry out more quickly in the summer and to be more easily replenished with water.

10. Grassing-down initially checked tree growth, especially in mixture A, in sub-plots with low nitrogen dressings, and in plots infrequently cut. Growth on the disced-in plots where checking had not been too severe caught up with growth on the cultivated plots. On the continued grass plots, except those receiving no added nitrogen, the growth rate recovered to normal and by 1946, in sub-plots of mixture B receiving 5 cwt./acre of sulphate of ammonia, exceeded that of the clean cultivated trees. All the trees, which at the outset of the experiment were growing poorly, looked well at the end of the period under review.

11. Grassing-down resulted in relative nitrogen shortage, as indicated by the pale leaf colour. A quick response was shown to added nitrogen, but leaf colour was as dark as that on the cultivated plots only when 10 cwt./acre of sulphate of ammonia was added. Discing-in of the sward resulted in a complete and immediate restoration of the dark green colour.

12. The amount of leaf scorches was slight, but there was some indication that their severity was less on the grass plots than on the cultivated area.

13. Grassing-down resulted in a marked reduction in pre-harvest dropping of fruit, this reduction being maintained even after discing-in the sward.

14. Fruit colour was greatly improved by grassing-down, but this improvement disappeared on discing-in the sward.

15. Where the swards greatly checked growth of trees, the crop was correspondingly reduced, but where the swards were suitably weak, adequately supplied with nitrogen and frequently cut, the crop was greater than on clean cultivated plots as well as of better quality. As a result of the reduced pre-harvest drop, all of the sward plots had a higher yield of picked fruit, in the second period of the experiment, than had the cultivated plots.

An increase of crop followed discing-in of vigorous swards after three years, but this did not quite make up for the severe check they had imposed while growing. In picked fruit, the plots three years with grass and subsequently four years with cultivation exceeded those cultivated throughout.

16. The presence of grass under the trees was found to make spraying, pruning and picking much easier. The use of a gangmower drawn by a rubber-tyred tractor, to keep the grass short, was very successful.

17. The experiment suggests that, under the soil and climatic conditions tested and with trees on rootstocks giving medium to vigorous growth, better results both in quality and quantity of crop may be attained by the use of weak growing swards, frequently cut, and receiving annual nitrogen dressings of the order of 5 cwt.

sulphate of ammonia per acre per annum, than by the use of clean cultivation with autumn weeds.

The importance of choosing the sward and treatment to suit soil, climate, rootstock and type of tree is emphasized.

ACKNOWLEDGMENTS.

The authors express their thanks to Dr. N. H. Pizer for carrying out the soil analyses in 1940, to Dr. E. W. Russell of Rothamsted Experimental Station for the loan of the compactometer, to Mr. F. H. Villiers for information regarding costs, to Mr. S. C. Pearce and his colleagues for the statistical analyses and to many members of the Pomology Section for carrying out much of the laborious work.

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(*Received* 3/9/48.)

COVER CROPS FOR FRUIT PLANTATIONS

V. EFFECT OF FORM AND TIME OF APPLICATION OF NITROGEN ON ORCHARD SWARDS

By **W. S. ROGERS, TH. RAPTOPOULOS** and **D. W. P. GREENHAM**

East Malling Research Station, Kent

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Introduction.

The inclusion of clovers or other legumes in swards used as orchard cover crops has been widely advocated, not merely for their value as pasture in grazed orchards, but also for the nitrogen which is fixed by their root nodules and presumably made available to the trees.

A number of workers have attempted to estimate the amount of nitrogen fixed by leguminous crops. Collison, Beattie and Harlan (1933) found that lucerne fixed 260 lb. of nitrogen per acre per year, equal to an annual application of 1,600 lb. of nitrate of soda. More recently, Chapman and Liebig (1947) have shown that purple vetch and *Melilotus indica* fixed about 1 cwt. of nitrogen per acre per year. In both cases the amount of nitrogen fixation was reduced when nitrogenous fertilizers were added. It has been claimed at the Pennsylvania Agricultural Experiment Station (1942) that under certain conditions orchards under leguminous swards can be self-supporting for nitrogen.

Investigations on the use of swards in apple orchards at East Malling (Papers I and IV of this series, Rogers *et al.*, 1945, 1948) have shown that unfertilized swards, even if completely or largely leguminous in composition, compete with the trees for nitrogen, especially in the early years of the sward, and tend to produce the characteristic nitrogen deficiency symptoms of poor leaf colour and reduced growth and cropping. If these symptoms are to be avoided, the use of nitrogenous fertilizers is necessary ; but it was found (Rogers *et al.*, 1948) that the addition of sulphate of ammonia resulted invariably in a rapid decrease of the clover content of the sward and in final disappearance of the clovers altogether. The same effect was found from the use of nitrate of soda in orchard rejuvenation trials in Ohio (Ballou and Lewis, 1920).

Much work has been done on the effect of nitrogenous manuring, grazing and shading on mixed swards by Blackman (1932a, 1932b, 1933, 1934, 1938) and by Blackman and Templeman (1938, 1940a, 1940b). In the first of these papers, reference is made to the work of Oakley (1925) who, in investigating methods of eliminating weeds from golf courses, found that nitrogenous fertilizers with an acid reaction, such as sulphate of ammonia, achieved this object while those with an alkaline reaction, such as nitrate of soda, encouraged the weeds. Oakley concluded that the degree of soil acidity induced by the fertilizers determined their effect on sward composition. Blackman (1932a) noted that sulphate of ammonia caused a decrease in the clover content of closely cut turf and later (1932b, 1934) he discounted the soil acidity hypothesis of Oakley and attributed the disappearance of clovers (amongst other weeds) to toxic effects of the ammonium ion ; he found nitrate of soda to have no effect on the sward composition. However, as a result of investigations on the interactions between nitrogen supply and light intensity on frequently cut agricultural swards, Blackman (1938) was forced to abandon the ammonium toxicity theory. Ammonia nitrogen was still found to reduce the clover content more than nitrate nitrogen and an alternative theory advanced (Blackman, 1938) involved assumptions of greater suppression of nodule formation by ammonia nitrogen or greater stimulation to growth of the grasses by ammonia nitrogen (Blackman, 1934). There was evidence from many workers that both nodule formation and nitrogen fixation are inhibited by a high external concentration of nitrogen, but others (e.g. Blackman and Templeman, 1938) showed that nitrogenous fertilizers did not depress the growth of clover alone, suggesting that the depression of clovers in swards by nitrogenous fertilizers is related to the presence of grasses. Blackman (1938) demonstrated that shading reduced the clover content and Blackman and Templeman (1938) reached the following general conclusion : " In grassland communities the balance between legumes and grasses is dependent largely upon competition for light. When the sward is infrequently defoliated then the taller growing grasses shade the clovers. Since the density and height of the grasses is correlated with the nitrogen supply, additional nitrogen may depress the leguminous species merely by increasing the degree of shading without any direct effect of nitrogen on the legumes."

In view of some of the evidence outlined above, it seemed possible that another nitrogenous fertilizer might be found more suitable than sulphate of ammonia for application on an orchard sward and this investigation was planned to compare the effects of several fertilizers.

Further, it was known that in a mixed grass and clover sward the grasses start to grow in the spring long before the clovers (Blackman, 1933), so it was thought that the time of application of nitrogenous fertilizers might well prove important in regulating the effect on sward composition. Consequently this trial was designed to include several nitrogenous fertilizers and several times of application, in an attempt to find an appropriate manurial treatment for supplying the fruit trees with adequate nitrogen and at the same time retaining the clovers in a mixed orchard sward.

MATERIAL AND METHODS.

Trees, soil and aspect.

The trial was carried out at East Malling during the years 1942 and 1943 in an orchard of bush Worcester Pearmain apples, part of which was used in the investigation of long-term leys in fruit plantations (Rogers *et al.*, 1948). As described in that paper, the plantation slopes gently down towards the north ; the soil is a rather shallow medium sandy loam with pH about 8, and is naturally rich in phosphates.

No detailed description of the trees is necessary here, as it was not the object of this investigation to observe any effects on them. It is sufficient to note that they were thirty years old and large enough to produce a fair amount of shade and to make considerable demands on the soil for water and nutrients.

Rainfall.

The average annual rainfall at East Malling for the thirty years 1915-1944 was 26·24 in. The actual totals for the years 1940-1943 were 27·01 in., 26·00 in., 26·25 in. and 25·70 in. respectively.

Seeds mixtures and sward management.

The experimental area was under the same two grass and clover mixtures, A and B, as were used in the investigation already mentioned (Rogers *et al.*, 1948). The seed was sown in the spring of 1940 and no fertilizer was applied until the start of this trial in 1942. The detailed composition of the seeds mixtures is shown in Table I. The main difference between the two mixtures is that A contained a large amount of cocksfoot and no red clover, while B included red clover but contained no cocksfoot.

TABLE I.

Composition of seeds mixtures.

	lb./acre.	
	A	B
Perennial ryegrass (*Lolium perenne*) (commercial)	8	20
Perennial ryegrass (*Lolium perenne*) (S.23) ..	6	—
Cocksfoot (*Dactylis glomerata*) (S. 143) ..	10	—
Rough stalked meadow grass (*Poa trivialis*) ..	2	4
Timothy (*Phleum pratense*)	—	4
Wild white clover (*Trifolium repens* var. *sylvestre*)	2	1
Late red clover (*Trifolium pratense* var. *serotinum*)	—	3
Broad red clover (*Trifolium pratense* var. *praecox*)	—	2

Mowing was carried out by a small motor-operated reciprocating grass cutter, the sward being cut four times in 1942 and three times in 1943, corresponding to the "infrequent" cutting treatment of paper IV, which was found to encourage grass at the expense of clover.

Experimental design.

A plan of the experiment is given as Fig. 1. The trees were in a diagonal plant 17 ft. apart. The width of the experimental area was 32 yd. and this was divided into two 16 yd. wide strips, one sown to each seeds mixture. The whole area was divided into three blocks as shown, each block containing 16 experimental treatments on each sward. The 16 plots in each half-block were of triangular shape, with a tree at each corner, the plot area being 16 square yards. The 16 treatments, whose positions were determined by randomization, consisted of 3 nitrogenous fertilizers × 5 times of application plus a control treatment receiving no manure. The total number of plots was thus 96.

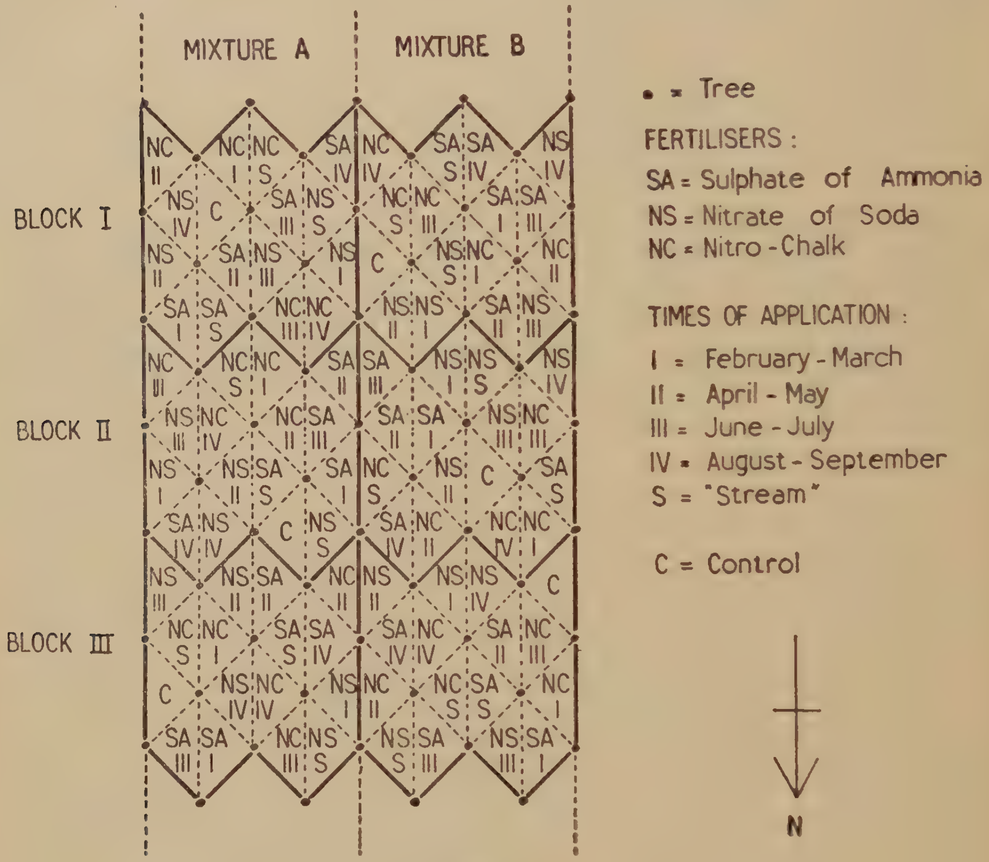
MIXTURE A
MIXTURE B
BLOCK I
BLOCK II
BLOCK III
• = Tree
FERTILISERS :
SA = Sulphate of Ammonia
NS = Nitrate of Soda
NC = Nitro-Chalk
TIMES OF APPLICATION :
I = February-March
II = April-May
III = June-July
IV = August-September
S = "Stream"
C = Control
N

Fig. 1.

Experimental design.

Manurial treatments.

The three fertilizers used were sulphate of ammonia, nitrate of soda and nitro-chalk. The rate of application of sulphate of ammonia was fixed at 4 cwt. (448 lb.) per acre per year and, in order that the same amounts of nitrogen should be given in all cases, equivalent amounts of the two other fertilizers were applied. Thus the amounts applied each year on each plot of 16 square yards were 1·5 lb. sulphate of ammonia, 2·0 lb. nitrate of soda or 2·0 lb. nitro-chalk.

The four times of application were February-March (I), April-May (II), June-July (III), August-September (IV), and in addition a "stream" (S) application was made, covering the whole period. In 1942 the total quantity of fertilizer was divided into two equal parts, one of which was applied in each of the two months of the appropriate treatment, but in 1943 the procedure was simplified by applying a single dose in one or other of the two months. In each year the stream application was made in four quarter-applications, one in each of the four periods.

The fertilizers were broadcast by hand, taking the experimental precaution of placing strings along all plot boundaries on each occasion. A dry day was always chosen for this operation, in order to avoid leaf scorch.

Botanical analysis.

The changes in sward composition were recorded by the percentage area method of Fenton (1933) as in the investigation described in paper IV of this series (Rogers *et al.*, 1948). In this method a 20 × 20 cm. square wooden quadrat sub-divided by thin wires into one hundred 2 × 2 cm. squares was thrown at random ten times on each plot. In assessing the percentage area covered, four categories were used—grasses, clovers, weeds (all plants other than grasses and clovers) and bare spaces. It was realized that, in order to avoid errors due to one species overlying and hiding from view another species, the record should be taken either before the occurrence of excessive growth in the spring or immediately after mowing. In this investigation the former alternative was adopted and records were taken in March, 1942 (before the application of any fertilizers), in March, 1943, and finally in early April, 1944.

Estimation of yield of swards.

The effect of manurial treatments on the yield of the swards was measured by weighing samples of cut material from an area of one square yard from each plot immediately after mowing. Sub-samples were taken for determination of dry matter content by oven-drying at 100° C., and the remainder of each sample was redistributed over the appropriate area.

RESULTS.

Effect on the sward composition.

Table II shows the percentage composition of the swards, in four categories, in 1942, 1943 and 1944. The data in this table represent the mean of all plots receiving nitrogen, compared with the mean of all control plots receiving no manure.

TABLE II.

Effect of nitrogenous fertilizers on the percentage composition of the swards.

	Nitrogenous fertilizers.				Control (no manures).			
	Grasses.	Clovers.	Weeds.	Bare spaces.	Grasses.	Clovers.	Weeds.	Bare spaces.
Mixture A.								
10th March, 1942	89·2	8·9	0·0	1·9	79·8	15·2	0·0	5·0
24th March, 1943	87·0	7·1	0·1	5·8	63·8	34·3	0·4	1·5
3rd April, 1944	84·3	2·0	0·0	13·7	60·1	31·2	0·0	8·7
Mixture B.								
10th March, 1942	67·5	28·7	0·0	3·8	73·5	23·7	0·0	2·8
24th March, 1943	84·5	11·2	0·3	4·0	63·8	29·0	1·0	6·2
3rd April, 1944	81·7	7·9	0·7	9·7	67·3	26·7	0·0	6·0

The application of nitrogen to sward A resulted in an increase in the proportion of bare spaces at the expense of both grass and clover but, since the proportion of clover was initially low, the effect on the grass was relatively unimportant compared with that on the clover which was reduced in two years to less than a quarter of its original amount. In contrast, on sward A receiving no manures, the clover, encouraged no doubt by the high natural phosphates, increased to over 30 per cent. at the expense of the grass, while the proportion of bare spaces was less than that on the plots receiving nitrogen. The contrast is illustrated in Figs. 2 and 3 which are photographs of sward A taken at about the same time as the final herbage analysis of 1944. Fig. 2 shows that the application of nitrogen resulted in a sward consisting almost entirely of cocksfoot, while much clover can be seen in the control sward (Fig. 3). The clover in sward A was entirely wild white.

The ratio of clover to grass was initially much higher in mixture B than in mixture A (Table II) but the same effect of nitrogen is seen. The amount of clover was reduced in two years almost to a quarter of its original proportion, whereas the initially high clover content was maintained in the absence of manures. Again, the proportion of bare spaces increased during the two years in presence or absence of fertilizers but not to the same extent as in the tufty cocksfoot of sward A. Figs. 4 and 5 illustrate the contrast in clover content in sward B. As in sward A, the clover here was entirely wild white; although red clovers were sown in mixture B, they were naturally dying out in 1942 (two years after sowing) and had quite disappeared by 1944.

In all cases the proportion of weeds was negligible, while the increase in the amount of bare spaces, although rather more marked in the presence of added nitrogen, occurred even when no nitrogen was added. This increase in the amount of bare spaces is probably related to the infrequent cutting treatment, as large masses of cut grass left to rot on the surface tend to smother the vegetation. Of the four categories the changes in the proportion of clover are the most important and these alone will be considered in greater detail.

TABLE III.

Effect of different types of nitrogenous fertilizers on percentage of clover in the swards.

	Sulphate of ammonia.	Nitrate of soda.	Nitro-chalk.	Control (No manure).
Mixture A.				
10th March, 1942 ..	9·3	9·3	8·2	15·2
24th March, 1943 ..	6·9	7·0	7·3	34·3
3rd April, 1944 ..	2·5	1·1	2·3	31·2
Mixture B.				
10th March, 1942 ..	28·0	27·3	30·8	23·7
24th March, 1943 ..	9·2	11·5	12·9	29·0
3rd April, 1944 ..	8·7	6·4	8·5	26·7

Table III shows the effect of the three different types of fertilizer on the clover content of the sward. The entries in each column represent the mean of all the corresponding " times of application " treatments. It is clearly seen that, for both mixture A and mixture B, the decrease in the percentage of clover was independent

PLATE I.

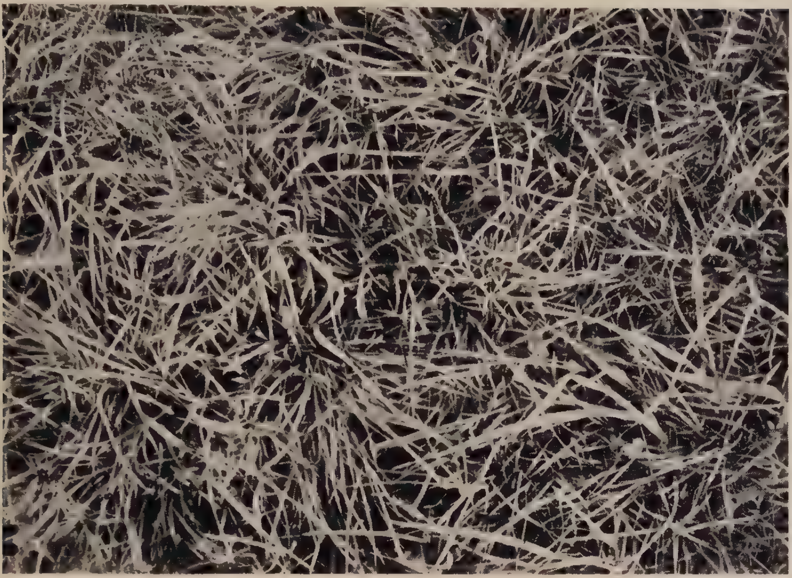

FIG. 2.
Sward A after two years "stream" application of sulphate of ammonia.

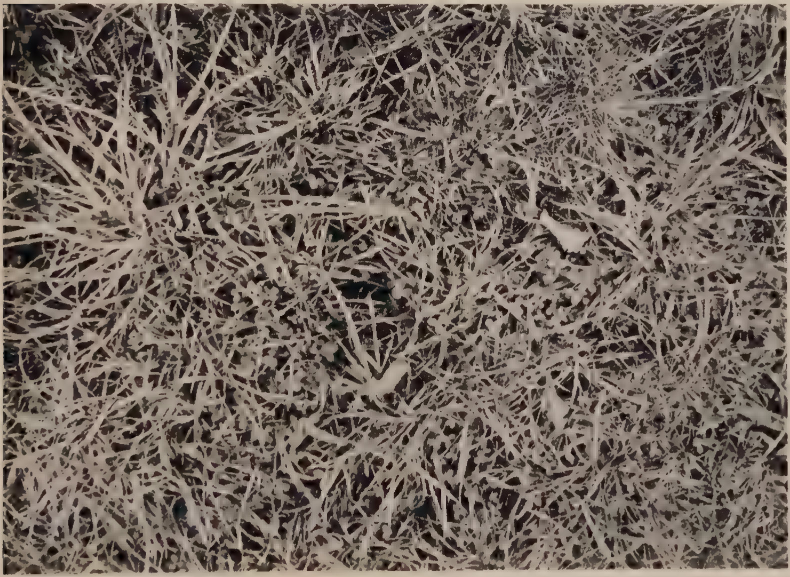

FIG. 3.
Sward A without fertilizers.

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PLATE II.

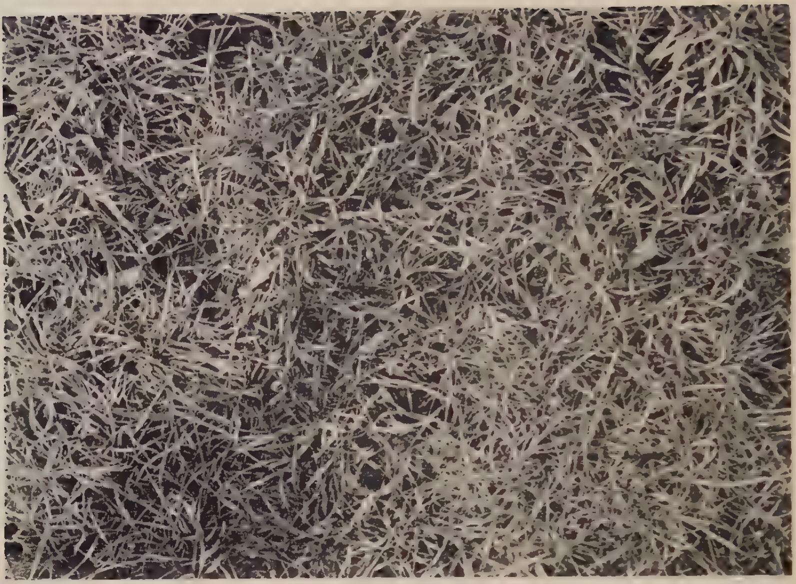

FIG. 4.
Sward B after two years " stream " application of sulphate of ammonia.

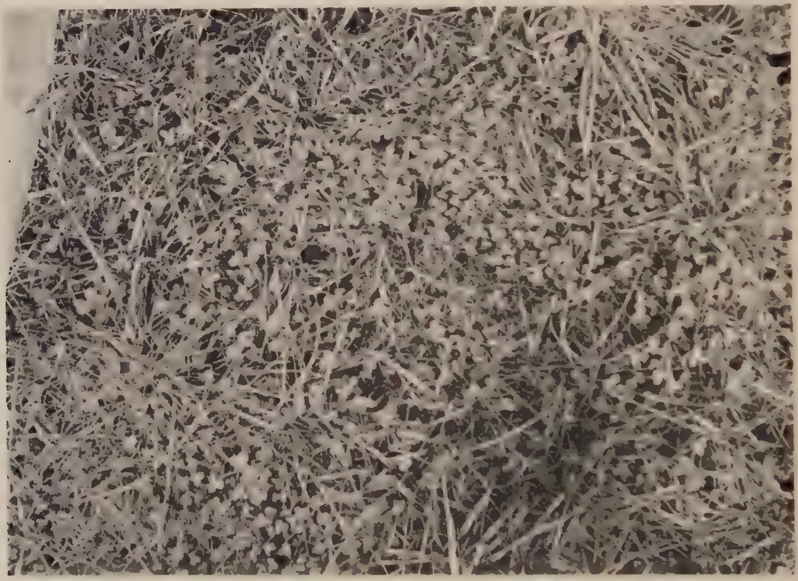

FIG. 5.
Sward B without fertilizers

of the particular form in which the nitrogen was applied. Considering the 1944 data, there were no significant differences between types of fertilizers but the percentage of clover in the control plots was significantly higher.

TABLE IV.

Effect of time of application of nitrogenous fertilizers on percentage of clover in the swards.

	I Feb.-March.	II April-May.	III June-July.	IV August-Sept.	S "Stream" application.	Control (No manures).
Mixture A.						
10th March, 1942 ..	8·9	7·6	7·9	11·4	8·9	15·2
24th March, 1943 ..	12·4	6·9	3·5	7·8	4·8	34·3
3rd April, 1944 ..	3·3	1·4	1·9	1·8	1·3	31·2
Mixture B.						
10th March, 1942 ..	26·8	31·0	27·2	32·7	25·8	23·7
24th March, 1943 ..	12·5	11·0	10·4	14·3	7·9	29·0
3rd April, 1944 ..	6·0	8·4	8·7	11·5	4·7	26·7

Table IV shows the effect of the different times of application on the clover content of the sward, all three types of nitrogenous fertilizer being considered together. For mixture A, the clover content, low initially, was reduced by all treatments to such very low proportions by 1944 that none of the differences in clover content in that year was significant, except that the control plots contained significantly more clover than the others. However, for mixture B, which included initially a much higher proportion of clovers, there were significant differences between times of application and these depended on the type of fertilizer. The times of application are arranged below in ascending order of clover percentages, and figures enclosed within brackets are not significantly different from each other :

Per cent. clover in swards, 1944.

	S.	I.	II.	IV.	III.
For sulphate of ammonia	4·5	6·2	7·1	9·6	16·1
	I.	S.	III.	II.	IV.
For nitrate of soda	3·3	3·7	4·8	7·6	12·7
	III.	S.	I.	II.	IV.
For nitro-chalk	5·3	5·8	8·6	10·6	12·1

The control plots contained 26·7 per cent. of clover in 1944, which is significantly greater than under any treatment.

So far as it is possible to generalize for all three types of fertilizer, it would appear that both early (I) and stream (S) applications are likely to have a more damaging effect on the clover content than an application as late as August-September (IV). It is interesting to note, however, that the June-July application (III) of sulphate of ammonia caused significantly less reduction in clover content than did any other time of application while for both nitrate of soda and nitro-chalk it had a significantly more harmful effect than did the later application (IV).

Effect on the sward yield.

Table V shows the sward yield in tons per acre, fresh and dry weight, with the corresponding dry matter content. The figures are the mean of all plots receiving nitrogen and the mean of all plots receiving no manures; they represent the total weight of vegetation cut throughout each of the seasons 1942 and 1943. As shown in the results of paper IV in this series (Rogers *et al.*, 1948), the application of nitrogen (in this instance 4 cwt./acre of sulphate of ammonia or its equivalent in the other fertilizers) had a marked stimulating effect on the total growth of the sward. This is true whether dry weight or fresh weight be considered, in fact the absence of any effect of nitrogen on the percentage dry matter content makes it justifiable to consider only the fresh weight figures in the detailed comparison of individual treatments.

TABLE V.

Effect of nitrogenous fertilizers on the sward yield.

	Nitrogenous fertilizers.			Control (No manures).		
	Fresh weight (tons/acre).	*Dry matter content* %	Dry weight (tons/acre).	Fresh weight (tons/acre).	*Dry matter content* %	Dry weight (tons/acre).
Mixture A.						
1942	11·2	*23·4*	2·6	8·1	*21·4*	1·7
1943	6·8	*27·3*	1·9	5·8	*28·4*	1·7
Total	**18·0**	*24·9*	**4·5**	**13·9**	*24·5*	**3·4**
Mixture B.						
1942	15·0	*20·1*	3·0	13·0	*21·1*	2·7
1943	8·0	*28·0*	2·3	5·4	*28·1*	1·5
Total	**23·0**	*22·9*	**5·3**	**18·4**	*22·8*	**4·2**

TABLE VI.

Effect of different types of nitrogenous fertilizers on sward yield (tons fresh weight per acre).

	Sulphate of ammonia.	Nitrate of soda.	Nitro-chalk.	Control (No manure).
Mixture A.				
1942	10·7	11·5	11·4	8·1
1943	7·3	7·0	6·1	5·8
Total	**18·0**	**18·5**	**17·5**	**13·9**
Mixture B.				
1942	15·5	14·1	15·4	13·0
1943	8·2	7·5	8·4	5·4
Total	**23·7**	**21·6**	**23·8**	**18·4**

Table VI shows the effect of the three different types of fertilizer on the sward yield. The figures represent the mean of all the corresponding " times of application " treatments. After each application the effect on the grass was noticeable in about a week but the response was rather more delayed in the case of sulphate of ammonia than with the other fertilizers. The yields are similar for all three types of fertilizer, whether mixture A or mixture B be considered. Statistical analysis shows that the slight differences between types of fertilizer are not significant but that all gave significantly higher yields than did the unmanured plots.

In both years the first cut, in May, and the subsequent cuts were made whenever the grass had reached a height of about 12 in. In 1942, the plots which were to receive their fertilizers in June-July (III) and in August-September (IV) were not sampled at the first cutting. In estimating the total yield from these treatments (Table VII) it has been assumed that the undetermined amounts were the same as the yield from the control (unmanured) plots. Since both were unmanured at the time, this assumption is reasonable for the purpose of compiling the table of total yields but treatments III and IV cannot be included in the statistical analysis.

TABLE VII.

Effect of time of application of nitrogenous fertilizers on sward yield (tons fresh weight per acre).

	I Feb.-March.	II April-May.	III June-July.	IV Aug.-Sept.	S "Stream" application.	Control (No manure).
Mixture A.						
1942	11·9	9·7	11·6	11·4	11·5	8·1
1943	9·7	6·4	5·0	5·5	7·6	5·8
Total	**21·6**	**16·1**	**16·6**	**16·9**	**19·1**	**13·9**
Mixture B.						
1942	15·6	16·0	14·5	14·1	15·0	13·0
1943	9·5	8·0	6·2	7·5	8·9	5·4
Total	**25·1**	**24·0**	**20·7**	**21·6**	**23·9**	**18·4**

Table VII is a comparison of the yields corresponding to different times of application, considering all three types of fertilizer together. For mixture A, the total yields for the two seasons together for treatments I, II, S and Control were all significantly different from each other, in the order I > S > II > Control. Treatments III and IV, which could not be considered in the analysis, each gave total yields approximating to that for treatment II. Thus, while nitrogen applied at any time increased the yield, the increase was greatest with the earliest application and almost as great with the stream application. For mixture B, none of the differences, except that between control plots and all the remainder, is significant, but the order of the yields is similar to that for mixture A: I > II > S > IV > III > Control and confirms the conclusion that early and stream applications exert a greater influence than do late applications of nitrogen on the growth of the sward.

It was shown in paper IV of this series (Rogers *et al.*, 1948) that most of the sward growth occurred before the end of May; the present results show that the high productivity at this peak period can be increased by the early application of nitrogen. Observations on the plots showed that, as noted above, the effect of nitrogen on the sward was appreciable about a week after application and that there was little or no residual effect after about two months. The exact length of that period was apparently related to the rainfall at the time but, after about two months from the time of application, the rate of growth was comparable with that on the unmanured plots.

DISCUSSION AND CONCLUSIONS.

The results of this investigation have shown that all three nitrogenous fertilizers used (sulphate of ammonia, nitrate of soda and nitro-chalk) had the same effect on mixed grass and clover swards. Use of each of these fertilizers resulted in an increase in the amount of fresh material produced and a decrease in the proportion of clovers in the sward.

The lack of any difference between the effects caused by nitrate of soda or nitro-chalk and by sulphate of ammonia does not confirm the results of Blackman (1932b, 1934, 1938) on clover content of swards, nor the observations of the same author on the growth of grasses (1934). But the swards in this investigation were allowed to grow long between cuttings, whereas Blackman's swards were always cut frequently. It is considered that the results of this investigation may be satisfactorily explained by the deleterious effects on the clover of increased competition by the grasses whose growth is stimulated by added nitrogen. This competition may have been for light, as concluded by Blackman and Templeman (1938). On the other hand, as discussed fully by Blackman (1938), nodule formation and nitrogen fixation appear to be inhibited by added nitrogen and, under those conditions when the legume is dependent upon external sources of nitrogen, it is unable to absorb nitrogen as rapidly as the grasses, so that competition for nitrogen may then be the determining factor in the relative proportions of grass and legume. Further, the stimulating effect of nitrogen being greater on grasses than on clover (Blackman and Templeman, 1938) may, as suggested by Blackman (1938), result in increased output of CO_2 by the grass roots or in increased secretion of some harmful substance, either of which may adversely affect nitrogen fixation. Whatever the mechanism, whether it be a shading effect or not, it seems probable that the decrease in clover content in the swards in this investigation may be attributed to increased competition by the grasses stimulated by application of nitrogenous fertilizers. This is in agreement with the general conclusion of Blackman and Templeman (1938), quoted in the introduction to this paper, relating to conditions in infrequently cut swards, except that this investigation provides no evidence regarding the mechanism of competition.

The biggest stimulation to the growth of the sward occurred when the fertilizers were applied early, before or during the period of maximum growth of the grasses. Since wild white clover makes most of its growth later in the season, one might have expected that these early fertilizer applications would not have had such a deleterious effect on the clover content of the sward. In actual fact, when the sward contained a high proportion of clover initially (sward B) the reverse effect was observed, i.e. the early applications caused the greatest reduction in clover content (although the June-July application of nitro-chalk was also found to be harmful). On any theory involving *direct* effect of fertilizers on clover, it is the later applications, at the time of maximum growth of the clovers, which would be expected to have the greatest suppressive effect on the clovers. Since the early applications also resulted in the greatest increase in sward yield, this may be regarded as evidence in favour of the theory that suppression of the clovers is caused indirectly by the fertilizers through their effect on the growth of the grasses ; the fact that the fertilizer is applied before the period of maximum growth of the clover itself makes no difference and it seems that the clover is probably suppressed before it has started to grow. When the clover content was low initially (sward A), the increased yield resulting from early application was even more marked, there being more grass to respond, but fertilizers applied at any time were sufficiently potent almost to eliminate such a small amount of clover ; the relationship between increased yield and suppression of clover cannot therefore be demonstrated in the case of sward A.

Fertilizers applied at intervals throughout the season February-September (" stream " application) were almost as efficient in increasing sward yield as when the whole amount was applied in February-March and were quite as potent in suppressing the clovers.

While greater reductions in clover content resulted from early applications of nitrogen, in the case of mixture B, the difference between early and late applications, though statistically significant, was not large enough to be of primary importance. The fruitgrower will naturally give first consideration to his trees rather than to his clover and will apply nitrogen early in the season, when it is most likely to be needed for tree growth. The resulting disappearance of clover appears at first sight to be inevitable, but, if it be considered desirable to maintain a high proportion of legumes in the sward, there are several possible ways in which this may be achieved :

(1) It may be possible, by injecting the nitrogenous fertilizer in solution into the soil, to ensure an adequate nitrogen supply for the tree without affecting the sward. A preliminary small-scale trial of this method is being undertaken at East Malling but it may not prove to be commercially practicable even if effective.

(2) It seems possible that the clover content of a mixed sward may be maintained, even in the presence of added nitrogen, if the sward is cut frequently. It is well known (Blackman, 1933) that hard grazing during the mid-season maintains or increases the clover content of pasture and it was shown in the previous paper of this series (Rogers *et al.*, 1948) that the disappearance of clover was less rapid when the sward was cut about six times a year than when cutting was infrequent (3 or 4 times a year), as in this investigation. These results incidentally support the theory that the clover is suppressed when the grass is encouraged to grow vigorously, in this case by infrequency of cutting. Further, it was shown in the same paper, from consideration of the behaviour of the trees, that frequent mowing is necessary. With the general adoption of the gangmower by fruit growers using swards, it is now common practice to mow even more frequently (10-12 times a year) than in that investigation, and, although experimental evidence on the effect of very frequent mowing in orchards is lacking, it may well be that the clover content can thereby be maintained. Under such conditions, the indirect effect of nitrogenous fertilizers acting through competition by the grasses will be reduced to a minimum. It is not known, however, whether any direct effect, no longer masked by the magnitude of the indirect one, would then be apparent ; for instance, sulphate of ammonia might then have a result different from that of nitrate of soda, as found by Blackman (1932b, 1934, 1938) for frequently cut swards.

(3) Probably the most practicable method of ensuring a continued high proportion of legumes, despite the heavy applications of nitrogen which appear to be necessary in the initial stages, is to include a much higher proportion in the seeds mixture, or even to sow a pure legume. This has not yet been tested experimentally but growers have reported favourable results from sowing pure wild white clover or the Aberystwyth strain S.100 of white clover. Natural grasses, such as *Poa* and *Agrostis*, can be expected to invade such a sward but their competing effect on the clovers can be minimized by frequent mowing. If invasion by grasses becomes too severe, the sward can be disced and resown periodically. Under such a system of management, the sward, although at first " locking up " nitrogen, may eventually fix and release sufficient nitrogen for the maintenance of the trees at a satisfactory level of growth and cropping.

(4) Although no experimental evidence is available on the relative effects of organic and inorganic nitrogenous fertilizers on sward composition, the possibility should be mentioned here that fertilizers such as meat and bone meal, releasing nitrogen slowly over a long period and supplying phosphate, may be less severe in their effect on the clover.

SUMMARY.

1. Equivalent amounts of inorganic nitrogen were applied in three different forms (sulphate of ammonia, nitrate of soda and nitro-chalk) to infrequently mown swards obtained by sowing two different grass and clover mixtures in an apple plantation. The fertilizers were applied at four different times (February-March, April-May, June-July, August-September) and by a " stream " application, consisting of a quarter-dressing at each of the four periods.

2. All plots with added nitrogen showed a decrease in the amount of clover, when compared with the control plots. All three forms of nitrogenous fertilizer were equally effective in suppressing the clover. When the clover content was initially low, almost complete suppression resulted from applications made at any time, but in swards containing much clover originally, the greatest reduction in clover content generally occurred when the fertilizers were applied early.

3. The yield of cut material obtained from the swards was considerably increased by the application of nitrogenous fertilizers, the increase being the same for all three forms of nitrogen. The greatest increase in yield resulted from the earliest applications.

4. As all three types of fertilizer were equally effective in suppressing clover, the effect was clearly not due to different reaction or to toxicity of any particular ion. The results can be interpreted most satisfactorily by an encouragement of the grass by application of nitrogen, resulting in " crowding out " the clover.

5. As the appearance of nitrogen deficiency symptoms in the early stages of " grassing down " orchards can only be avoided by the application of nitrogenous manures, and as an inevitable result of such applications is the elimination of clovers from the usual grass and clover mixtures, it is suggested that the most likely method of obtaining a sward capable of supplying nitrogen to the trees is to sow a pure legume such as wild white clover and minimize the competing effect of any invading grasses by frequent mowing.

ACKNOWLEDGMENT.

The authors express their thanks to Mr. S. C. Pearce and his colleagues for the statistical analyses.

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(*Received* 27/9/48.)

A SUPPLEMENTARY NOTE ON FRUIT SPRAYING WITH A GLYOXALIDINE PREPARATION IN 1948

By **R. W. MARSH**
Long Ashton Research Station, Bristol

In a previous paper in this Journal (Marsh, 1947) promising results were recorded from spraying trials using heptadecylglyoxalidine and hydroxyethyl heptadecylglyoxalidine for the control of apple Scab and black currant Leaf Spot. As stated in that paper, the results were open to the objection that the trials were made during a dry summer. These results can now be supplemented by those available for the more normal summer of 1948. The relevant 1948 figures for monthly rainfall and days in which measurable rain was recorded at Long Ashton were: April, 2·22 in. (14); May, 3·67 in. (11); June, 2·59 in. (21); July, 1·34 in. (13); August, 4·69 in. (17).

PLOTS AND MATERIALS.

Apples. The apple spraying trials were again carried out on Plot 23 of the Long Ashton plantations. For the Scab control trial, the 13-year-old Worcester Pearmain trees in sub-plot N were used. These stood originally at 15 ft. on the square, but were thinned in the winter of 1947-48 to a diagonal plant at approximately 21 ft. (see Fig. 1). Trials to investigate the possibility of spray damage were made on adjacent blocks of Cox's Orange Pippin of the same age and spacing.

Black currants. The black currants used for spraying trials were, as in 1947, the Baldwin block on Plot 3 at Long Ashton, fully described by Marsh and Dickinson (1944).

Materials. A fluid preparation of glyoxalidines was provided by the Carbide and Carbon Chemicals Corporation (N.Y.). One quart of this material was said to contain 1 lb of a mixture of 2-heptadecylglyoxalidine, 2-heptadecenylglyoxalidine and 2 pentadecylglyoxalidine. For convenience this preparation will be referred to here as Glyoxalidine 341 CA. It was prepared for use by diluting 1 quart to 100 gallons with water and adding 1 lb. of hydrated lime.

EXPERIMENTAL DATA.

Apples.

Scab Control Trial.

The layout for this trial is shown in Fig. 1. Trees lettered A were sprayed with lime sulphur at 2 per cent. on 13th April, 2 per cent. on 23rd April, 1 per cent. on 15th May and 1 per cent. on 10th June. At each of these dates the trees lettered B were sprayed with Glyoxalidine 341 CA at 1 quart/100 gallons (plus 1 lb. hydrated lime). Trees lettered C were given no fungicidal spray but, in common with the rest of the plot, received a nicotine-Estol H spray on 12th May for the control of Sawfly.

The pattern of replication was designed to ensure that any spread of Scab infection from tree to tree in the path of the prevailing S.W. wind would be retained within the same treatment series.

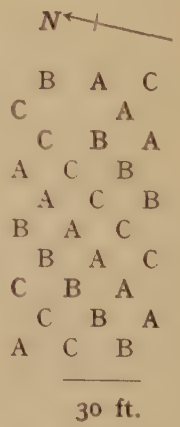

Fig. 1.

All spraying was done at low pressure, using hand-operated nozzles to minimize drift; the rate of application was approximately

2 gallons per tree. The second pre-blossom spray was carried out in gusty weather; otherwise, the conditions for spraying were satisfactory.

Scabbed leaved were first seen on the control trees on 15th May and the disease spread steadily in June and, more slowly, in July. A 250-leaf sample was taken from each tree in the last week in July and the mean percentage scabbed area determined as described by Kearns, Marsh and Martin (1945).

In the last week of August, all the fruits from each tree were picked and the percentage scabbed area recorded, using the diagram given by Marsh (1947). Results are shown in Table I. At the same time the percentage number of fruits with serious russeting was noted for each tree (see Table II).

TABLE I.

Results of apple Scab control trial.

Treatment.	Mean percentage surface area scabbed.	
	Per leaf.	Per fruit.
Lime Sulphur	0·602	0·106 n.s.
Glyoxalidine 341 CA	0·116	0·058
No fungicide	1·112	0·309

(All differences except that marked n.s. are significant at $P < 0{\cdot}01$.)

It will be noted from Table I that the mean percentage leaf infection on the glyoxalidine-sprayed trees is only one-tenth of that on the unsprayed; on the fruit the proportion is approximately one-fifth. On the leaf counts, but not on the fruit, the glyoxalidine treatment gave significantly better Scab control than the standard lime sulphur programme.

Phytotoxicity trials.

(*a*) *On Worcester.* From July onwards, russeting of fruit became noticeable in the Worcester block referred to above. Counts of all fruits, made at the time of picking, gave the results shown in Table II.

TABLE II.

Mean percentage numbers of fruits showing disfiguring russet.

Sprayed with lime sulphur.	Sprayed with glyoxalidine.	Control.
8·5	9·2	13·1

These figures showed no significant differences.

(*b*) *On Cox.* A block of 120 trees of Cox's Orange Pippin on Plot 23 was sprayed with lime sulphur at 2 per cent. on 15th April, 1 per cent. on 12th May and 1 per cent. on 10th June. On each of these dates an adjoining block of 30 trees of Cox was sprayed with Glyoxalidine 341 CA at 1 quart per 100 gallons (plus 1 lb. hydrated lime). All treatments were carried out with a P40 power sprayer using 400 lb.

pressure and 3 nozzle brooms fitted with 6/64 in. discs. Drenching applications were made at a rate of not less than 3½ gallons per tree. It was impracticable to lay out a replicated trial because of spray drift problems.

From May onwards the foliage of all Cox plots at Long Ashton in 1948, irrespective of treatment, showed much wind damage and " Cox spot ". The fruit crop on this variety was light throughout the plantations.

Slight leaf fall was noted in mid-May on the glyoxalidine-sprayed blocks and very slight leaf fall on the lime sulphur-sprayed. After the June spraying, the position was reversed and the leaf fall on the glyoxalidine-sprayed Cox's was very slight, while that on the lime sulphur-treated blocks was clearly greater. However, none of the damage attributable to either spray treatment in this trial would be considered of commercial importance. The fruits harvested from the experimental plots showed no spray damage and were completely free from Scab.

BLACK CURRANTS.

In the earlier paper (Marsh, 1947) the results were given of the 1947 trial using heptadecylglyoxalidine against black currant Leaf Spot. In 1948 the same plot of Baldwin black currants was used and divided into three blocks, within each of which the following spray treatments were randomized on 14-bush replicates : Phygon (dichlornaphthaquinone) 1 lb/100 gallons ; Glyoxalidine 341 CA 1 quart/100 gallons plus 1 lb. hydrated lime ; pentachlornitrobenzene (20 per cent.) 5 lb./100 gallons ; no spray.

The sprays were applied on 26th June at a rate of 1⅓ gallons per bush, using a power sprayer and spray brooms. At this date the fruit was half ripe and *Pseudopeziza* attack on the leaves was just starting.

The fungus developed slowly in 1948 and the final count was not made until 14th October. At that date the mean percentage foliage retention for the four treatments was :

Control.	*Pentachlornitrobenzene.*	*Phygon.*	*Glyoxalidine.*
26	29	54	74

These figures confirm the 1947 findings, that glyoxalidine provides an effective control of the Leaf Spot caused by *Pseudopeziza ribis.*

The fruits were picked on 13th July and taken to the Fruit and Vegetable Preservation Research Station at Campden for processing trials.

Results from these trials are not yet available but those from the previous year's crop have been reported by Adam, Dowson and Marsh (1947). These showed that the residue from a glyoxalidine spray on the currants canned in 1947 did not act as an accelerator of corrosion and had no adverse effect on the flavour or appearance of the processed fruit.

ACKNOWLEDGMENTS.

The author is indebted to the Carbide and Carbon Chemicals Corporation and to Messrs. Bayer Products for spray materials, to Miss C. E. C. Clarke, Mr. S. H. Bennett and Mr. A. H. Fielding for carrying out the spraying operations and also to the last-named for recording results.

SUMMARY.

In a season favourable to apple Scab attack, spraying Worcester Pearmain trees four times with a preparation of glyoxalidines reduced the amount of leaf infection by 90 per cent. and of fruit infection by 80 per cent. compared with that on unsprayed trees. On the leaves, the glyoxalidine treatment provided significantly better Scab control than that given by the standard lime sulphur spray programme.

The glyoxalidine sprayings resulted in no damage to the foliage or fruit of the Worcester Pearmain trees. Three heavy sprayings with the glyoxalidine preparation caused negligible foliage damage on Cox's Orange Pippin.

The value of glyoxalidine for the control of *Pseudopeziza ribis* on black currant was confirmed.

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Marsh, R. W. (1947). Fruit spraying trials with certain recently introduced fungicides. J. Pomol., **23**, 185.

(*Received* 27/10/48.)

REVIEWS

COPPER COMPOUNDS IN AGRICULTURE AND INDUSTRIAL MICROBIOLOGY. C.D.A. Publication No. 41, 1948, 117 pp., 46 Figs. Copper Development Association, Grand Buildings, Trafalgar Square, London, W.C.2., and Kendals Hall, Radlett, Herts.

The author has performed a useful service in searching out from existing literature the information contained in this book. The result of his task is a handy reference work compiled with evident care.

He has oriented his survey of the microbiological field, largely concerning plant diseases and their control, along a copper thread, which is not without piquancy for the mycologist or plant pathologist or allied chemist, accustomed to classification of a different kind. If occasionally, as in the chapter on spray machinery, the thread seems strained to its limit, the liberty is permissible in the general pattern.

Much additional information is contained in chapters on copper insecticides, seed treatment, weed control, copper deficiency diseases, copper as an antiseptic, and the uses of copper in industrial microbiology (textile and timber preservation, and the treatment of water " scums ").

The book is indexed and has a bibliography of the principal works consulted. Attractively produced and liberally illustrated with some fine reproductions, it is available free on application to " those giving evidence of their genuine interest in the subject ". M.H.M.

OUTDOOR SALAD CROPS. Bulletin No. 55 of the Ministry of Agriculture and Fisheries. (London : His Majesty's Stationery Office, 1948. Pp. 42. Price 1s. net.)

This is the fourth edition of a bulletin first published in 1932. It is written for the market grower and has nothing to do with kitchen garden methods and problems. Eleven of its pages are devoted to lettuce, and the other crops dealt with at varying length are endive, chicory, radish, celery, spring onions, corn salad, ridge cucumbers, tomatoes and watercress. As can be judged from the list, the treatment of some of these crops is necessarily brief, but by concise writing the bulletin packs a great deal of information into small compass and should be widely useful. E.E.C.

PLUMS AND CHERRIES. Bulletin No. 119 of the Ministry of Agriculture and Fisheries. Pp. vi + 74. 16 Plates. (London : His Majesty's Stationery Office, 1948. Price 2s. 6d. net.)

This publication deals in detail with the growth conditions, management factors, and routine cultural operations necessary for the successful commercial cultivation of plums and cherries. The information is set out under the same general headings as in Bulletin No. 133 on Apples and Pears, and this uniformity of treatment has much to commend it. For each fruit separate sections are included on growth conditions, tree form and rootstock, varieties, planting systems, cultivations, grass management, pruning, spraying, picking and marketing. The lists of market varieties are short, concise, and up to date and, together with the tables of blossoming times, they make it possible for the intending planter to plan his list of varieties in the most suitable order for cross-pollination and for successional picking. Readers who are accustomed to the sober prose of Ministry publications will be delighted with Mr. Robert Hart's lyrical breakaway on page 57 where he describes the birds hiding in the twiggy windbreak " when the scarer passes on his rounds ".

No bulletin can be absolutely perfect, and the defects in this one might equally be the result of having too many editors or none at all. The inclusion in two bulletins, this one and No. 133, of almost identical paragraphs and illustrations of subjects such as Planting Boards and Planting Systems seems unnecessary and so does the inclusion of information on the same subject, e.g. soil and climate, in two successive chapters, instead of its being neatly combined in one paragraph in one chapter. As an intending planter of plums, one could not help feeling confused, after reading on page 1, " on the whole the finest and longest lived trees are found on medium to deep loams", to find on reaching page 3 that " there seems little doubt that plum trees thrive best on heavy loams or calcareous clays ". N.B.B.

PUBLICATIONS RECEIVED.

EMENDATIONS TO THE THIRD EDITION OF THE LIST OF COMMON BRITISH PLANT DISEASES. By a Sub-committee of the Plant Pathology Committee of the British Mycological Society. (Reprinted from The Transactions of the British Mycological Society, Vol. XXXI, Parts III and IV, 1948).

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